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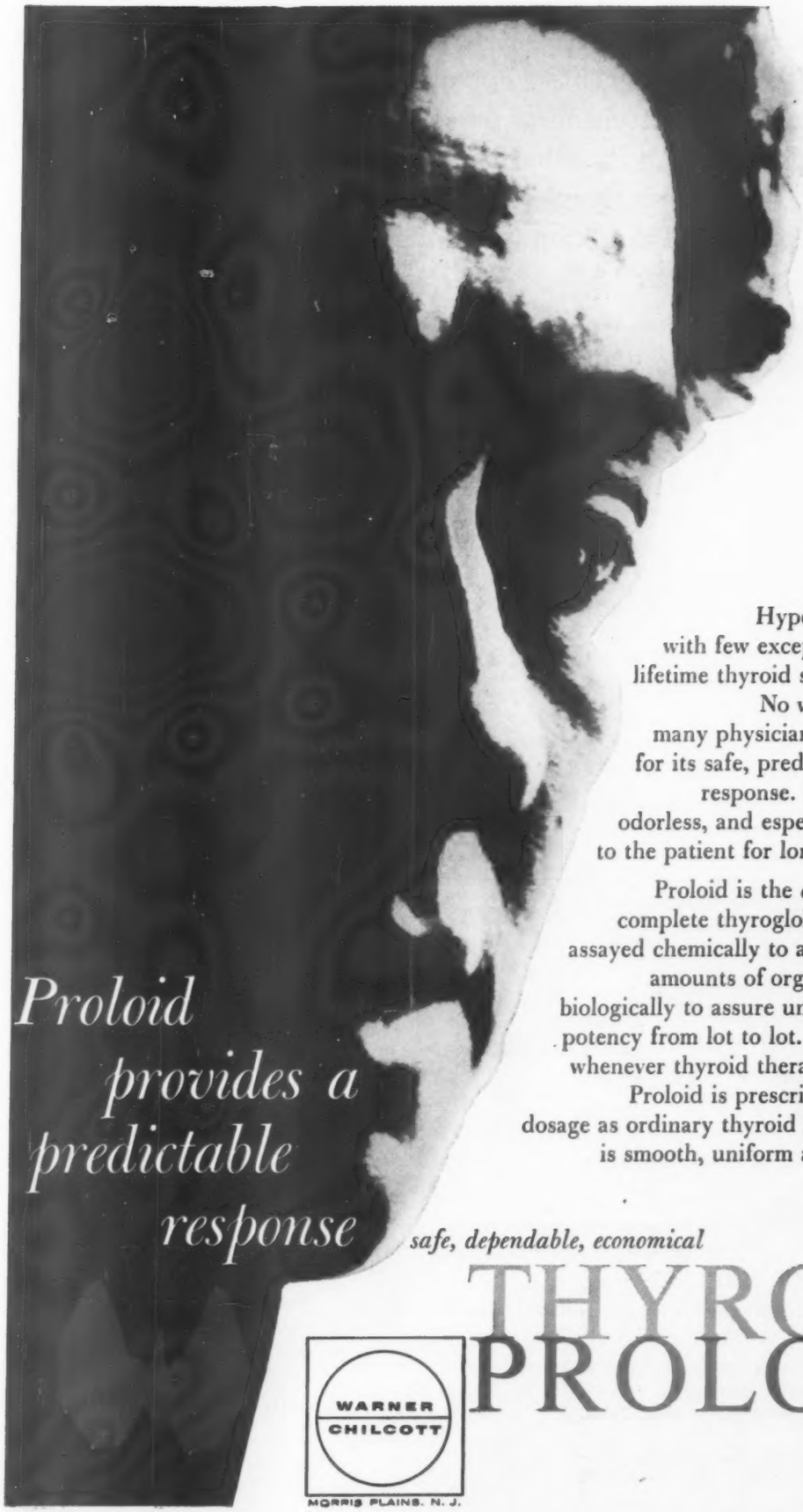

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1. Whitelaw, J.W.: Hypoparathyroidism in Children. *Bull. Vancouver Med. Assn.*, 30:116, Dec., 1953.
2. Sandock, Isadore: Tetany and Ovarian Function. *J.A.M.A.*, 160:659, Feb. 25, 1956.
3. Grollman, Arthur: Essentials of Endocrinology. Philadelphia, J. B. Lippincott Co., 2nd ed., 1947, p. 269.

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This fine study of leptospirosis in its milder forms, a somewhat neglected subject in this country, deserves attention. From the twelve cases reported and others cited it is clear that the classic full blown picture characterized by fever, jaundice, renal damage and hemorrhage is the exception in leptospiral infection, which is more likely to be manifested by headache, myalgia, fever, chills, conjunctival suffusion and, after the first week of illness, by aseptic meningitis in exposed patients. Dr. Edwards makes the further point that the association of any characteristic symptomatology with any specific leptospiral serotype is apt to be misleading, the clinical pictures overlapping too much. It is suggested that the diphasic course of most leptospiral infections is attributable to an initial leptospiremia followed by manifestations reflecting the host's immune response to the organisms.

Asian Influenzal Pneumonitis. A Structural and Virologic Analysis
PETER J. SOTO, JR., GORONWY O. BROUN AND JOHN P. WYATT 18

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Studies on the Origin of Drug-Resistant Staphylococci in a Mental Hospital
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In order to study the emergence of drug-resistant staphylococcal strains in a hospital environment the investigators repaired to a mental hospital in which antibiotics had been infrequently employed and systematically studied the effect of administration of penicillin, tetracycline and erythromycin. Despite the infrequent prior usage of these antibiotics, strains resistant to penicillin and tetracycline soon appeared in large numbers; the experience with erythromycin was more encouraging, but resistant strains also ultimately emerged. Susceptible strains rapidly disappeared. Perhaps the best policy, the authors conclude, is to reserve the use of erythromycin, kanamycin and other agents effective in the control of staphylococcal infections specifically for such infections.

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(1) Rubin, A., and Babbott, D.: J.A.M.A. *168*:498, (Oct. 4) 1958. (2) Kinsey, A. C.; Pomeroy, W. B., and Martin, C. E.: Sexual Behavior in the Human Male, Philadelphia, W. B. Saunders Company, 1948.

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The authors describe a peculiar form of jaundice occurring late in pregnancy, disappearing promptly upon delivery, and recurring with subsequent pregnancy. The clinical, laboratory and liver biopsy manifestations all indicate a form of intrahepatic biliary tract obstruction, with cholestasis, of unknown pathogenesis. The course is benign except for the threat of hemorrhage. A strong case for differentiation from viral hepatitis, jaundice associated with toxemia of pregnancy, drug hypersensitivity jaundice and other known forms of jaundice in pregnancy is made. Further study of this syndrome will be awaited with interest.

Biochemical, Blood Gas and Peripheral Circulatory Alterations in Hepatic Coma
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Additional data are presented to support the association (and presumptive causal relationship) of increased arterial blood ammonia levels and impaired consciousness in patients with liver disease, particularly evident when individual patients are studied serially in progressive cerebral dysfunction. Peripheral A-V ammonia differences correlated significantly with the arterial blood ammonia level and the peripheral A-V oxygen differences. Other changes studied in patients with hepatic coma of progressive intensity included respiratory alkalosis, decreased oxygen saturation of arterial blood, and azotemia, all of which evidently are the result of multiple causes interacting in complex ways. It is made clear that simultaneous measurement of different parameters gives a more comprehensive picture of the scope of the abnormality in hepatic coma than does any single study.

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Vitamin B_6 Deficiency and Oxalate Nephrocalcinosis in the Cat
STANLEY N. GERSHOFF, FAROUK F. FARAGALLA, DOUGLAS A. NELSON AND STEPHEN B. ANDRUS 72

It is shown that diets grossly deficient in vitamin B_6 cause an interesting oxalate nephrocalcinosis in cats, in addition to the more generally recognized deficiency pattern. What, if any, implications this may have for the genesis of hyperoxaluria and oxalosis in man is not yet clear. However, in view of the quite confused state of knowledge, certainly of prevention and treatment of such conditions in man, the observation deserves attention and further study.

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1. Nussbaum, H. E., Leff, W. A., Mattia, V. D., Jr. and Hillman, E.: Am. J. M. Sc. *234*:150, Aug. 1957. 2. Dunsmore, R. A., Dunsmore, L. D., Bickford, A. F. and Goldman, A.: Am. J. M. Sc. *233*:280, March 1957. 3. Boyd, L. J., Huppert, V. F., Mulinos, M. G. and Hammer, H.: Am. J. Cardiol. *3*:229, Feb. 1959.

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Erythrocyte Cholinesterase Titers in Hematologic Disease States JEAN C. SABINE 81

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Seminar on Mycotic Infections

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S. B. SALVIN 97

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References: 1. Crunk, G. A.; Naumann, D. E., and Casson, K.: Antibiotics Annual 1957-1958, New York, Medical Encyclopedia Inc. 1958, p. 397 • 2. Newcomer, V. D.; Wright, E. T., and Sternberg, T. H.: Antibiotics Annual 1954-1955, New York, Medical Encyclopedia Inc., 1955, p. 686.

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Advertising Index on Page 131

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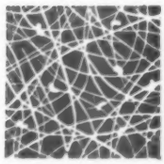
the use of vitamins

in rheumatoid arthritis



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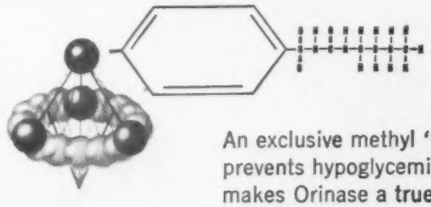

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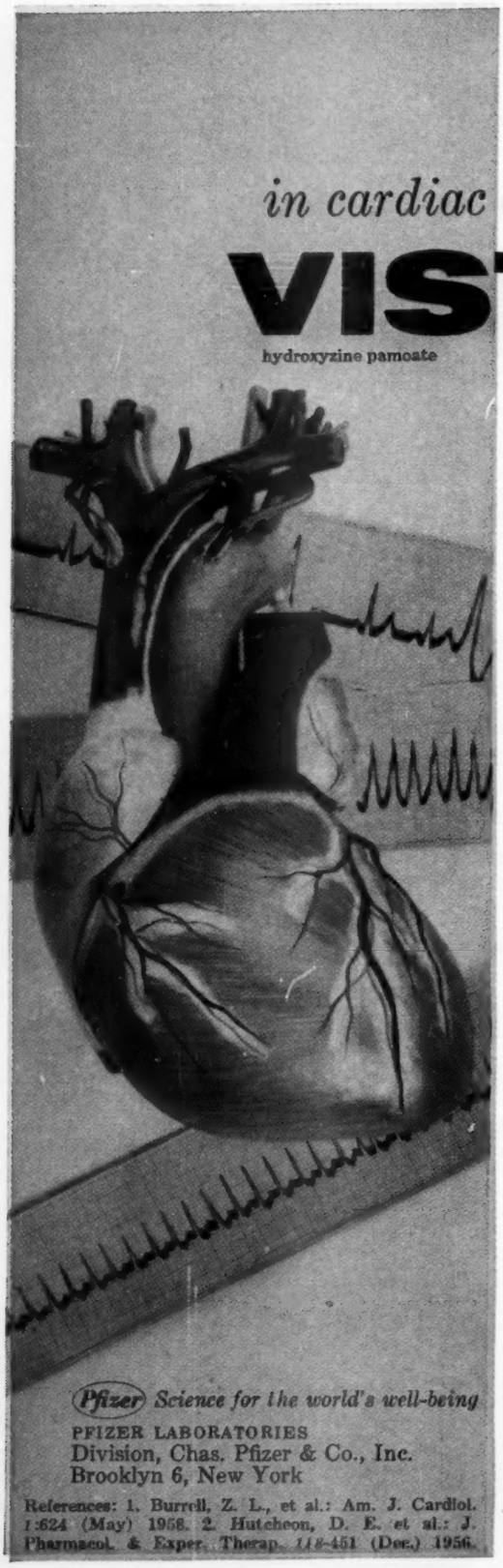

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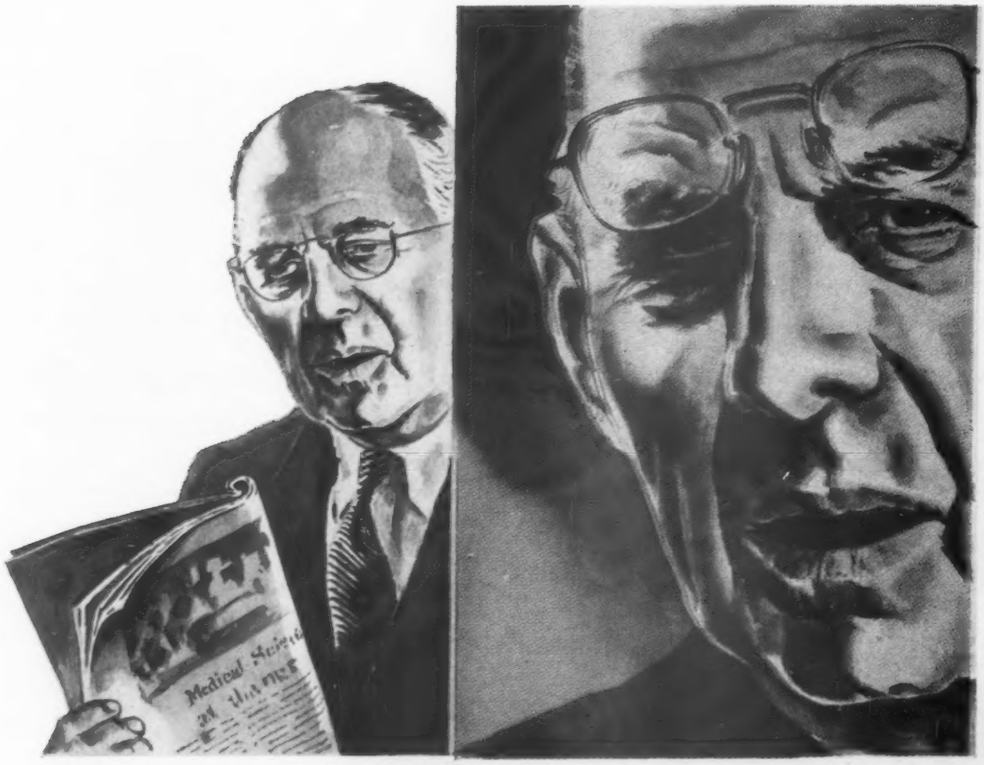

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Physicians who have used PROTAMIDE extensively deplore such statements as unfortunate when they appear in the lay press. They have repeatedly observed in their practice quick relief of pain, even in severe cases, shortened duration of lesions, and greatly lowered incidence of postherpetic neuralgia when PROTAMIDE was started promptly. A folio of reprints is available. These papers report on zoster in the elderly—the severely painful cases—patients with extensive lesions. PROTAMIDE users know "shingles" *can* be helped.

PROTAMIDE®

Sherman Laboratories

Detroit 11, Michigan

Available: Boxes of 10 ampuls—prescription pharmacies.

TO BE SPECIFIC, DOCTOR

for nausea • vomiting • vertigo BONAMINE*

brand of meclizine hydrochloride

Clinically proved relief up to 24 hours with a single dose

BONAMINE (a non-phenothiazine) is remarkably free of side effects

BONAMINE HAS NO KNOWN CONTRAINDICATIONS–CAN BE USED WITH CONFIDENCE FOR AS LONG OR AS OFTEN AS REQUIRED

BONAMINE *Tablets,* scored, 25 mg. Boxes of 8, bottles of 100 and 500.

BONAMINE *Chewing Tablets,* pleasantly mint flavored, 25 mg. Packages of 8.

BONAMINE *Elixir,* cherry flavored, 12.5 mg. per 5 cc. Bottles of one pint.

DOSAGE: Usually 25 to 50 mg. once a day.

PFIZER LABORATORIES Division, Chas. Pfizer & Co., Inc. Brooklyn 6, N.Y.

*TRADEMARK

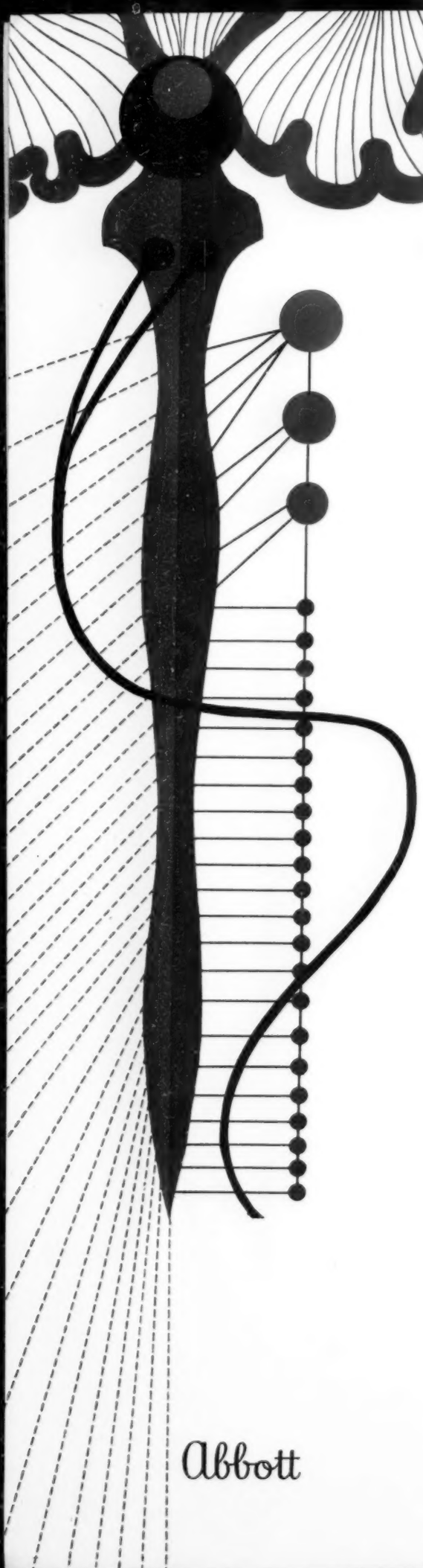
Science for the world's well-being Pfizer

Mild and persuasive as a lullaby,
nonbarbiturate Placidyl gently lulls your
patients into refreshing slumber.
Brief and effective.
Prescribe it this week and see.

Abbott

Placidyl® nudges your patient to sleep

(Ethchlorvynol, Abbott)



and the evidence mounts

"Blood pressure in the hypertensive patient was lowered gently and not as precipitously as with reserpine."[1]

"In our total experience with this drug (deserpidine), no significant side effects were observed requiring discontinuation of the medication."[2]

"We found deserpidine to be a drug of low toxicity and well-tolerated by practically all of our patients."[3]

"While the results were qualitatively similar to those with reserpine in similar dosage ranges, side-effects were much less frequent, very mild, and none severe enough to interrupt treatment."[4]

". . . In contrast to our experience with reserpine, we found a greater degree of safety with deserpidine and relatively lower toxicity. Also, it was better tolerated by our patients."[5]

"Side effects were notably absent in patients treated with Harmonyl."[6]

". . . side reactions appear to be less annoying and somewhat less frequent." (with deserpidine).[7]

". . . R canescens (base of Harmonyl) tended to have fewer and less severe side effects. In particular it seemed to have less sedative effect than R. serpentina."[8]

1. Frohman, I. P., Tranquilizers in General Practice and Clinical Evaluation of Deserpidine, an Alkaloid of Rauwolfia canescens, M. Ann. District of Columbia, 27:641, December, 1958.

2. Rawls, W. B., et al., Clinical Experience with Deserpidine in the Management of Hypertension and Anxiety Neurosis, New York J. Med. (in press).

3. Billow, B. W., et al., The Use of a New Rauwolfia Derivative, Deserpidine, in Mild Functional Disturbances and Office Psychiatry, New York J. Med. (in press).

4. Ibid.

5. Billow, B. W., et al., The Effect of a New Rauwolfia Derivative, Deserpidine, in Hypertension, New York J. Med., 58:3641, November 15, 1958.

6. Ferguson, J. T., Comparison of Reserpine and Harmonyl in Psychiatric Patients, A Preliminary Report, Journal-Lancet, 76:389, December, 1956.

7. Moyer, J. H., et al., Deserpidine (Canescine) for the Treatment of Hypertension, South. M. J., 50:499, April, 1957.

8. Achor, R. W., and Hanson, N. O., Hypertension Treated with Rauwolfia canescens. A Comparison with Rauwolfia serpentina, New England J. Med., 255:646, October, 1956.

filmtab® **HARMONYL®**

(Deserpidine, Abbott)

for your next working hypertensive

Abbott

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What's Your Corticosteroid Score?

corticosteroid quiz

		True	False
1	Therapeutic corticosteroid action may be explained by the inhibition of antigen-antibody reactions.	☐	☐
2	The enhanced potency of the "Meti" steroids is partly due to a slower metabolic rate.	☐	☐
3	Corticosteroids can alter the triple response to intradermally administered histamine.	☐	☐

See below for answers.

scores highest in clinically important tests

METICORTEN®
prednisone

Unsurpassed effectiveness with minimal mineralocorticoid activity—proved in practice. These are the observations confirmed in hundreds of published papers by clinical investigators using METICORTEN in the most complete variety of steroid-responsive disorders.

METICORTEN—1, 2.5 and 5 mg. tablets.
Meti,® brand of corticosteroids.

Schering

SCHERING CORPORATION • BLOOMFIELD, NEW JERSEY

answers: 1. False—corticosteroids act by inhibiting tissue response to cellular injury. 2. True. 3. False—generally corticosteroids do not alter cutaneous reactions that develop rapidly and are associated with the release of histamine-like compounds.

S-201

Control of acute agitation: as close as this

You are always prepared to cope with acutely agitated patients when SPARINE is in your bag. SPARINE calms the patient quickly, reducing both the emotional and physical manifestations of agitation and apprehension.

The prompt control obtained with injectable SPARINE can be maintained by the use of SPARINE intramuscularly or orally.

Sparine® HYDROCHLORIDE

Promazine Hydrochloride, Wyeth

Injection **Tablets** **Syrup**

Wyeth®
Philadelphia 1, Pa.

If your patient has high blood pressure plus one or more of these complications:

anxiety
congestive failure
tachycardia
edema/overweight

control all the symptoms with just one prescription

new Esidrix-Serpasil®

(hydrochlorothiazide and reserpine CIBA)

Combination Tablets

new Esidrix-Serpasil:

B.P.: 180/125 mm. Hg
Heart rate: 96 beats/min.
Enlarged heart

High blood pressure plus tachycardia

Therapy: Esidrix-Serpasil. ***Rationale:*** Heart-slowing effect of Serpasil to prolong diastole, allow more time for recovery of myocardium, increase coronary blood flow, improve cardiac efficiency. Potentiated antihypertensive effect for greater blood pressure control.

B.P.: 205/145 mm. Hg
Orthopnea
Venous engorgement
Ascites

High blood pressure plus congestive failure

Therapy: Esidrix-Serpasil. ***Rationale:*** Potent diuretic action of Esidrix to relieve edematous condition, improve cardiac status. Combined antihypertensive action of Esidrix and Serpasil for lowest blood pressure levels. Convenience of combination tablet medication for greater patient acceptance.

one prescription that controls high blood pressure plus its complications

B.P.: 220/140 mm. Hg
Edema
Weight: 210 pounds

High blood pressure plus edema/overweight

Therapy: Esidrix-Serpasil. *Rationale:* Diuretic effect of Esidrix to eliminate excess body fluids, bring patient to dry weight. Potentiated antihypertensive effects of Esidrix and Serpasil in combination. Convenience of 1-prescription therapy.

B.P.: 170/112 mm. Hg
Nervous
Sweating palms

High blood pressure plus anxiety

Therapy: Esidrix-Serpasil. *Rationale:* Central action of Serpasil to calm the patient, shield him from environmental stress. Combined antihypertensive action of Esidrix and Serpasil for lowest blood pressure levels. Simplified dosage schedule.

one prescription that controls high blood pressure plus its complications

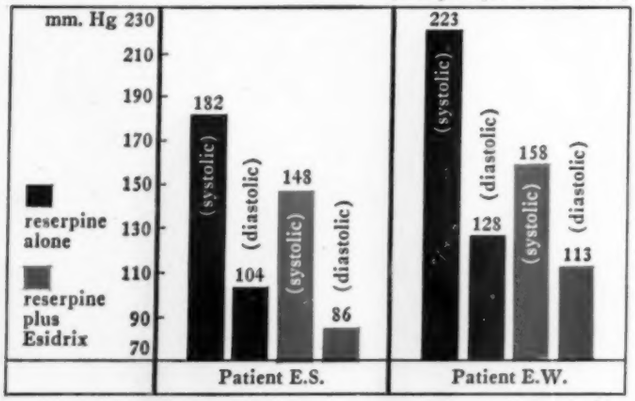
Esidrix-Serpasil Combination Tablets

A new antihypertensive combination—Esidrix-Serpasil is a combination of ESIDRIX[T.M.] (hydrochlorothiazide CIBA), an improved analog of chlorothiazide developed by CIBA research, and SERPASIL® (reserpine CIBA). Each tablet combines the potent diuretic and mild antihypertensive effects of Esidrix with the antihypertensive, heart-slowing and calming effects of Serpasil.

Indications—Esidrix-Serpasil is indicated in all grades of hypertension, particularly when one or more of the following complications exist: anxiety, tachycardia, congestive failure, pitting edema, edema of obesity, other edematous conditions.

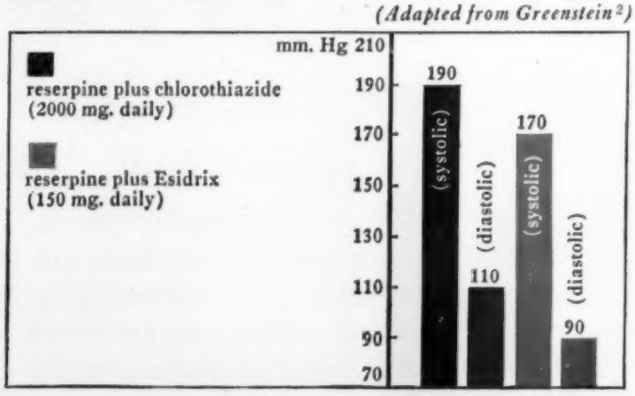
More effective than either drug alone—Investigators who have used the combination of hydrochlorothiazide and reserpine report that it is more satisfactory than either drug alone.

(Adapted from Maronde[1])



More effective than chlorothiazide-reserpine combinations—Many patients resistant to chlorothiazide-reserpine therapy have shown significant clinical response when Esidrix-Serpasil was started. The blood pressure of patient shown below was only slightly reduced on chlorothiazide and reserpine. When Esidrix was substituted for chlorothiazide, lower blood pressure levels were achieved.

(Adapted from Greenstein[2])



Dosage—Esidrix-Serpasil is administered orally in a dosage range of 1 to 4 tablets daily. Each tablet contains 25 mg. of Esidrix and 0.1 mg. of Serpasil. The total daily dose may be given after breakfast or in 2 or 3 divided doses. Dosage in every case should be individualized and adjusted to meet changing needs.

Since the antihypertensive effect of Serpasil is not immediately apparent, the maximal reduction in blood pressure may not occur for 2 weeks. At this time the dosage of Esidrix-Serpasil should be adjusted to the amount necessary to obtain the desired blood pressure response. For maintenance, as little as 1 tablet daily may be sufficient.

In cases of more severe hypertension, dosage of Esidrix-Serpasil can be revised upward to 4 tablets daily. When necessary, more potent antihypertensive agents such as Apresoline, Ecolid or other ganglionic blockers may be added. As Esidrix-Serpasil potentiates the action of other antihypertensive drugs, such additions to the regimen should be gradual and effects carefully observed. When Esidrix-Serpasil is started in patients already receiving ganglionic blockers, such as Ecolid, dosage of the latter should be immediately reduced by at least 50 per cent.

Side effects and cautions—As when any diuretic agent is used, patients should be carefully observed for signs of fluid and electrolyte imbalance. Esidrix in therapeutic doses is generally well tolerated. Side effects, even from large doses, have been few. Since Esidrix greatly reduces the amount of Serpasil needed, the incidence of side effects sometimes encountered with Serpasil is diminished.

Complete information on Esidrix-Serpasil available on request.

Supplied—Esidrix-Serpasil *Tablets,* 25 mg./0.1 mg., each containing 25 mg. of Esidrix and 0.1 mg. of Serpasil; bottles of 100.

References—1. Maronde, R. F.: Clinical Report to CIBA. 2. Greenstein, S.: Clinical Report to CIBA.

APRESOLINE® hydrochloride (hydralazine hydrochloride CIBA)
ECOLID® chloride (chlorisondamine chloride CIBA)

new Esidrix-[T.M.] Serpasil® (hydrochlorothiazide and reserpine CIBA)
Combination Tablets

CIBA
SUMMIT, N. J.

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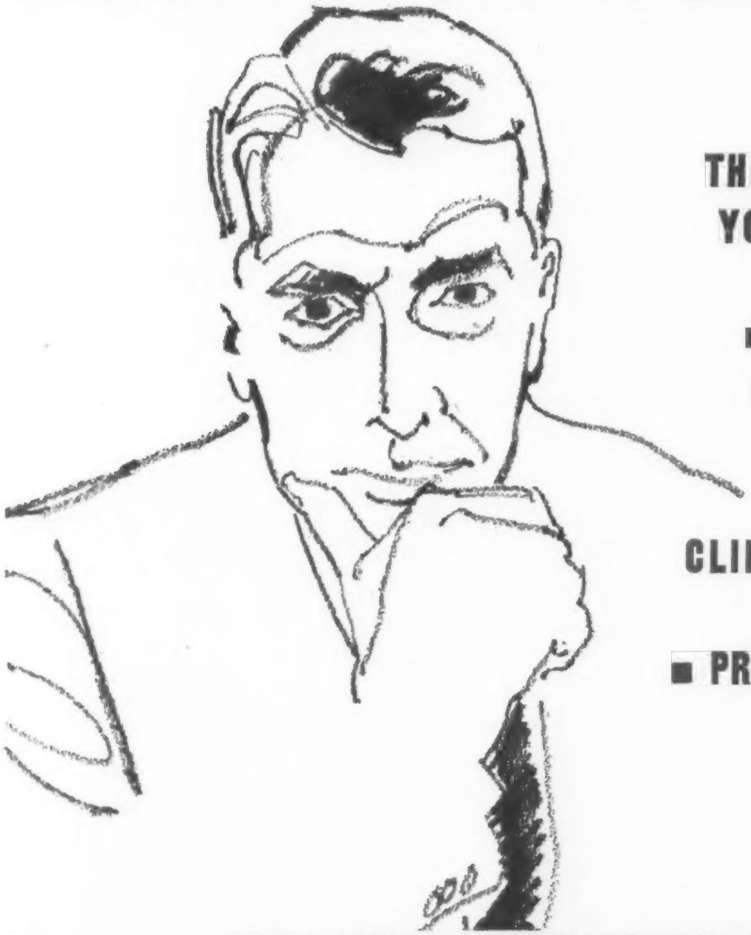
doctor, if you are not completely satisfied



...IF

THE ANTIHISTAMINES YOU ARE NOW USING:

- **ARE INEFFECTIVE IN SOME PATIENTS**
- **ARE LIMITED IN CLINICAL APPLICATION**
- **PRODUCE UNDESIRABLE SIDE EFFECTS**

THEN YOUR NEXT ALLERGIC PATIENT MAY NEED

New TOTAL ACTION **"THERUHISTIN" FORTE**

Brand of Isothipendyl hydrochloride

Sustained Action Tablet for daylong antiallergic effect

A new antihistaminic/anticholinergic/antiserotonin agent of "high potency and efficiency"[1] **sustained relief** in an average of over 90 per cent of 1209 cases of various allergic diseases[1-3] ■ **strikingly free of side effects** ■ **has been used successfully in all varieties of allergy**... of considerable value in bronchial asthma

Also available: "THERUHISTIN" Syrup. Potency recently increased from 2 to 4 mg. per 5 cc. (1 tsp.). *Usual dosage for children:* ½ to 1 tsp. two to four times daily depending on age.

Usual Dosage: One tablet morning and evening.

Supplied: No. 737—24 mg. Sustained Action Tablets, bottles of 100 and 1,000.

1. Committee on New and Unused Therapeutics, Am. Coll. Allergists: Ann. Allergy 16:237 (May-June) 1958. 2. Spielman, A. D.: Ann. Allergy 16:242 (May-June) 1958. 3. Spielman, A. D.: New York J. Med. 57:3329 (Oct. 15) 1957.

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NOW—YOU CAN PRESCRIBE THE UNSURPASSED ADVANTAGES OF

superior antiallergic efficacy with new low dosage

NEW

Arist

- combines the **anti-inflammatory, antiallergic** and **antihistaminic** effects of two agents—ARISTOCORT and chlorpheniramine which, separately, have been proved highly effective in the treatment of allergy
- permits **greater latitude** in adjusting dosage to minimum level needed for maintenance, because ARISTOCORT and chlorpheniramine are supplied in the **lowest dose tablets** available for each component alone
- supplies **ascorbic acid** for increased demand in stress conditions

Indications: *Generalized pruritus of allergic origin; hay fever, allergic rhinitis, perennial asthma, seasonal and perennial rhinitis, vasomotor rhinitis; drug reactions and other allergic conditions.*

Dosage: *One to eight capsules a day in divided doses. Dosages should be established on the basis of individual therapeutic response.*

Precautions: *Drowsiness may occur, and is usually due to the antihistamine effect. Occasionally this may also cause vertigo, pruritus and urticaria. Because of the low dosage, side effects with* ARISTOMIN *have been relatively infrequent and minor in nature. However, since* ARISTOCORT *Triamcinolone is a highly potent glucocorticoid with profound metabolic effect, all precautions and contraindications traditional to corticosteroid therapy should be observed. Discontinuance of therapy must not be sudden after patients have been on steroids for prolonged periods. It must be carried out gradually over a period of as much as several weeks.*

Further information available on request.

Supply: *Each* ARISTOMIN *Capsule contains:*

ARISTOCORT® Triamcinolone..... 1 mg.
Chlorpheniramine Maleate..... 2 mg.
Ascorbic Acid.....75 mg.

Bottles of 30 and 100

References: *1. Maurer, M. L.: Clinical Report, cited with permission. 2. Levin, L.: Clinical Report, cited with permission. 3. Gaillard, G. E.: Clinical Report, cited with permission.*

ARISTOCORT IN ANTIHISTAMINE COMBINATION

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Steroid-Antihistamine Compound LEDERLE

(lung x 65, injected with carbon-gelatin)

comments by clinical investigators:

"I would conclude that ARISTOMIN *is truly a worthwhile aid in treating allergic problems."*[1]

"The results have been uniformly good. The patients have stated that their symptoms were very much relieved. I have not encountered any side reactions except from one patient, who complained of some drowsiness, which I attribute to the antihistamine."[2]

"In general . . . it [ARISTOMIN] *is an excellent product. Over-all, it appears to be more effective than any simple antihistamine we have used. Despite the fact that we employed it in the treatment of a variety of nonselected individuals and problems, we had excellent and good results in 25 of the 39 patients."*[3]

Lederle

LEDERLE LABORATORIES, A Division of AMERICAN CYANAMID COMPANY, Pearl River, N. Y.

DR. MONTAGUE:
The first one I'll try your new obesity product on is YOU, Mr. Carr.

MR. CARR:
You've convinced me. I'll make the "one for good measure"
AMPLUS IMPROVED.

new AMPLUS® improved

(D-AMPHETAMINE + ATARAX® + VITAMINS AND MINERALS)

One capsule half-hour before each meal. Bottles of 100 soft, soluble capsules, this actual size.

DR. MONTAGUE AND MR. CARR DETAIL #2

DR. MONTAGUE: Well! Putting it on around the middle, aren't you?

MR. CARR: Wait a minute. This is the other side of my split personality. I'm not your patient today. I'm Joe Carr — Roerig detail man . . . remember?

DR. MONTAGUE: A lot's happened since I saw you last — about eight pounds, I should say. But have it your way, for the moment. What's on your mind?

MR. CARR: It's a new, improved product for — well — er — obesity.

DR. MONTAGUE: If I'm going to replace my present prescription for obesity, you'd better have a good reason.

MR. CARR: I have, Doctor. Three of them, in fact.

DR. MONTAGUE: I'll start counting.

MR. CARR: Well, AMPLUS has d-amphetamine to control the somatic factor, hunger pangs. And — stop looking at me that way.

DR. MONTAGUE: Sorry. Just a professional habit.

MR. CARR: Number two: Atarax — the tranquilizer that makes dieting endurable — with antisecretory action to stop that want-to-raid-the-icebox feeling.

DR. MONTAGUE: Go on. You fascinate me.

MR. CARR: I'm bloody but unbowed. Three! Vitamins and minerals to maintain nutrition during the critical period.

DR. MONTAGUE: I'll try it IMMEDIATELY.

MR. CARR: Thanks, Doctor. I'll be in for my usual checkup.

DR. MONTAGUE: Just a second. How about one for good measure.

MR. CARR: O.K. I'll play.

DR. MONTAGUE: I detect that high-calorie-coffee-break gleam in your eye. Make sure that "one for good measure" is AMPLUS IMPROVED.

MR. CARR: Now hold it! It's only temporary . . . A few rounds of golf . . . Remember, today I'm Joe Carr, Roerig detail man . . . I'll just leave you these samples . . .

DR. MONTAGUE: Here. Keep some for yourself. You've just been elected my *first* AMPLUS IMPROVED patient. And I've got a lot of other patients in mind for it. Good combination. Keeps appetite down, nutrition up, and certainly a tranquilizer is a fine idea in any regime as trying as dieting. And the antisecretory property of Atarax. That's a nice plus. . . .

MR. CARR: I hate to interrupt. But I have the strangest feeling that I've heard all this before.

representing

J. B. Roerig and Company

Mr. Carr

ROERIG Rx

New York 17, N. Y.
Division, Chas. Pfizer & Co., Inc.
Science for the World's Well-Being

ROCHE LABORATORIES IS PRIVILEGED TO ANNOUNCE A NEW DEVELOPMENT TO STOP AS WELL AS PREVENT VOMITING AND NAUSEA

Tigan

The first specific antiemetic/antinauseant entity

TIGAN IS NOT A CONVERTED ANTIHISTAMINE, NOT A CONVERTED TRANQUILIZER, NOT A CONVERTED SEDATIVE, NOT A COMBINATION

1

Chemically
different as well as new —
a specific antiemetic entity.[1]

2

Pharmacologically
different as well as new —
no demonstrable effects
other than antiemesis.[2]

3

Therapeutically
different as well as new —
stops *active* vomiting in
addition to prophylactically
preventing nausea and
emesis.[1,3-5]

4

Clinically
different as well as new —
effective against vomiting
and nausea in the widest
range of common and special
situations, such as
pregnancy, travel sickness,
gastrointestinal disorders,
uremia, carcinomatosis,
drug poisoning,
radiation sickness,
postoperative states.[1,3,4-6]

5

Practically
different as well as new —
patients may drive,
fly and work in hazardous
situations, even when
previously interdicted
with other agents.[1,2,4-6]

Controlled clinical investigations encompassing 3,000 patients have been completed to date.

No special precautions—
No known contraindications

Tigan affords such antiemetic efficacy, safety and pharmacologic precision that virtually all of the special precautions that have complicated older therapies are obviated. The singular absence of side effects makes it possible to use Tigan even in the presence of common contraindications of older antiemetics.[1-9]

Dosage: **The recommended adult dosage is one or two capsules four times a day.**

Available: **Capsules — 100 mg, blue-and-white, bottles of 100 and 500.**

References: 1. Reports on file, Roche Laboratories. 2. W. Schallek, G. A. Heise, E. F. Keith and R. E. Bagdon, *J. Pharmacol. & Exper. Therap.*, in press. 3. I. Roseff, W. B. Abrams, J. Kaufman, L. Goldman and A. Bernstein, *J. Newark Beth Israel Hosp.*, 9:189, 1958. 4. W. B. Abrams, to be published. 5. O. Brandman, to be published. 6. Personal communications. 7. P. K. Conner, Jr., S. Kinard, W. Fraser, H. Bennett and J. H. Moyer, Scientific Exhibit, A.M.A. Clinical Meeting, Seattle, Nov. 27-30, 1956. 8. *New and Nonofficial Drugs*, Philadelphia, J. B. Lippincott Co., 1959, p. 362. 9. J. H. Moyer, in *Drugs of Choice 1958-1959*, St. Louis, The C. V. Mosby Co., 1958, p. 336.

TIGAN[T.M.]—brand of trimethobenzamide ROCHE®

ROCHE LABORATORIES
Division of Hoffmann-La Roche Inc.
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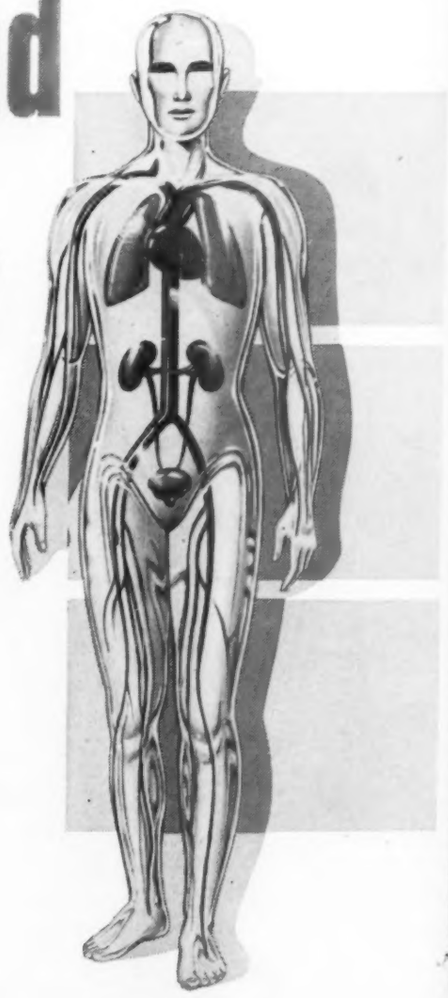
NEW

HYDRODIURIL™

(HYDROCHLOROTHIAZIDE)

simplifies* and improves any regimen for hypertension



*it's as easy as 1, 2, 3 to use

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(HYDROCHLOROTHIAZIDE)

1 Initiate therapy with HYDRODIURIL: one 25 mg. tablet or one 50 mg. tablet once or twice a day. HYDRODIURIL by itself often causes an adequate drop in blood pressure over a period of two to three weeks. This may be all the therapy some patients require.

2 Add or adjust other agents as required: HYDRODIURIL enhances the activity of all commonly-used antihypertensive agents; thus, the dosage of other medication (rauwolfia, reserpine, hydralazine, veratrum) should be initiated or adjusted as indicated by patient condition. If a ganglion-blocking agent is contemplated or being used, usual dosage must be reduced by 50 per cent.

3 Adjust dosage of all medication: the patient must be frequently observed and careful adjustment of all agents should be made to establish optimal maintenance dosage.

Supplied: 25 mg. and 50 mg. scored tablets HYDRODIURIL (Hydrochlorothiazide) bottles of 100 and 1,000.
Additional literature for the physician is available on request.

HYDRODIURIL is a trademark of Merck & Co., Inc. Trademarks outside the U. S.: DICHLOTRIDE, DICLOTRIDE, HYDROSALURIC.

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"To know how to grow old is the master-work of wisdom..."
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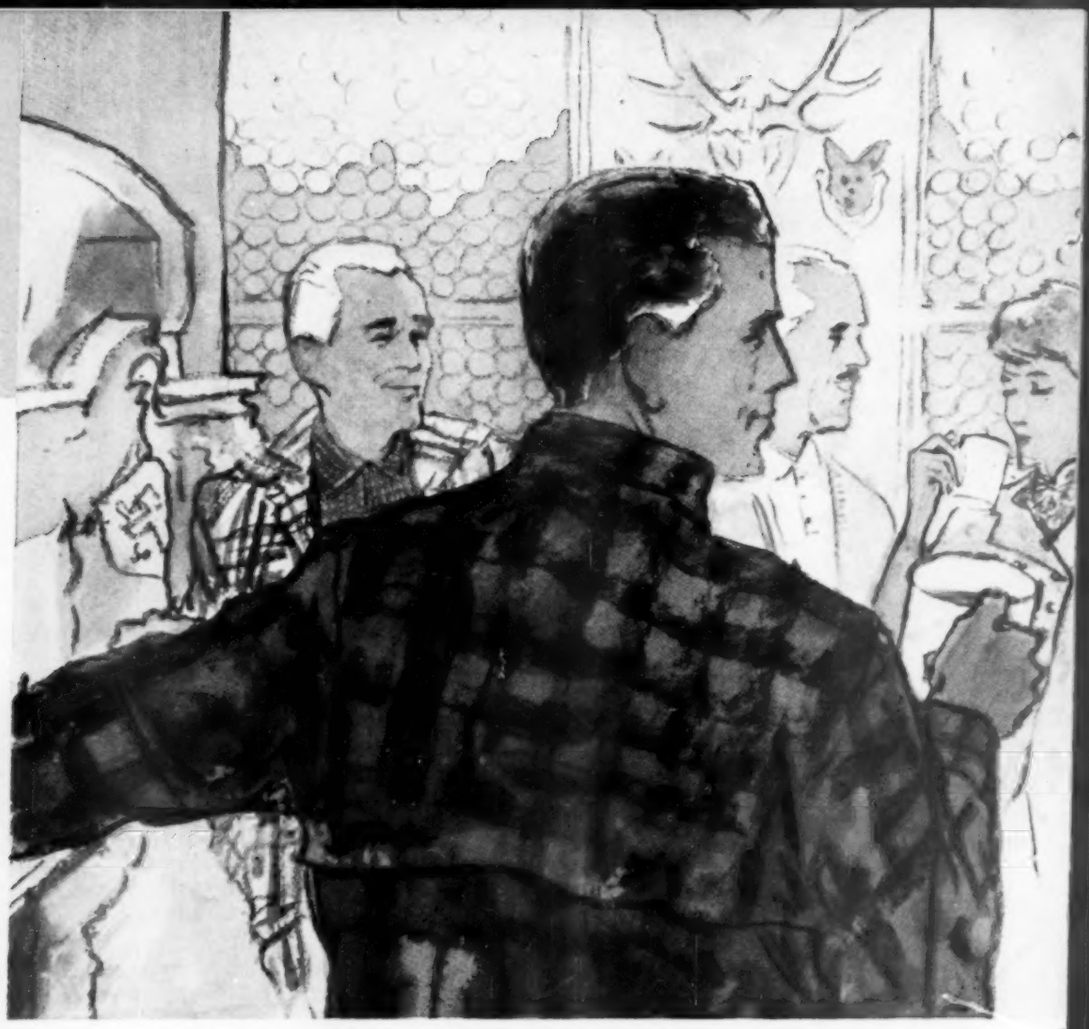

for your "over forty" patients prescribe

ELDEC® KAPSEALS®

vitamin-mineral-hormone supplement

to help fill their later years with vigor and vitality

each KAPSEAL *contains:*

vitamins	
Vitamin A	1,667 Units (0.5 mg.)
Vitamin B_1 mononitrate	0.67 mg.
Ascorbic acid	33.3 mg.
Nicotinamide	16.7 mg.
Vitamin B_2	0.67 mg.
Vitamin B_6	0.5 mg.
Vitamin B_{12} with intrinsic factor concentrate	0.033 USP Unit (oral)
Folic Acid	0.1 mg.
Choline bitartrate	6.67 mg.
Pantothenic acid (as the sodium salt)	5 mg.
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Ferrous sulfate (exsiccated)	16.7 mg.
Iodine (as potassium iodide)	0.05 mg.
Calcium carbonate	66.7 mg.
digestive enzymes	
Taka-Diastase® (Aspergillus Oryzae Enzymes)	20 mg.
Pancreatin	133.3 mg.
protein improvement factors	
l-Lysine monohydrochloride	66.7 mg.
dl-Methionine	16.7 mg.
gonadal hormones	
Methyl testosterone	1.67 mg.
Theelin	0.167 mg.

dosage: One Kapseal three times daily before meals. Female patients should follow each 21-day course with a 7-day rest interval.

packaging: ELDEC Kapseals are available in bottles of 100.

MEDICAMENTA VERA · PD & Co.

PARKE, DAVIS & COMPANY, Detroit 32, Michigan

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Remarkable relief from

LOW BACK PAIN

and

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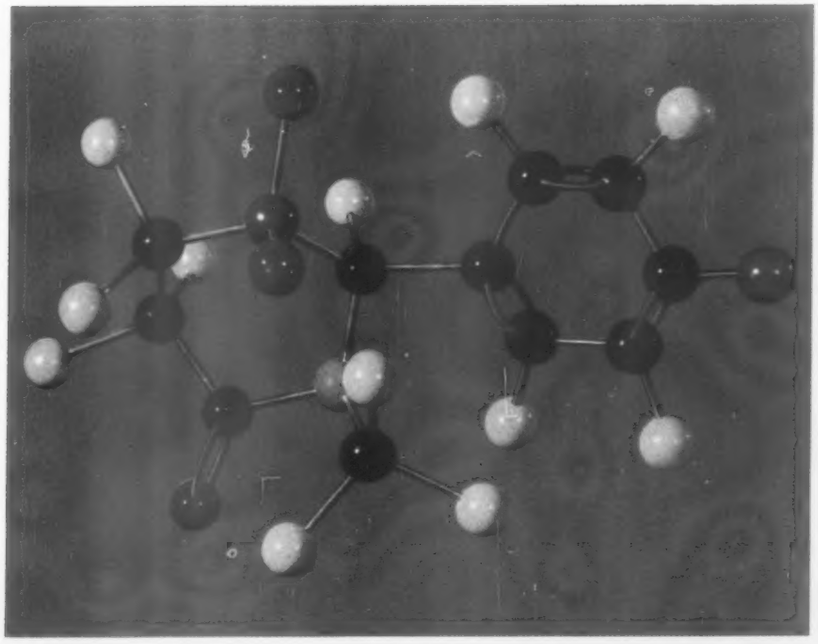
Trancopal®

*the first true tranquilaxant**

***Potent* MUSCLE RELAXANT**

***...Equally effective as a* TRANQUILIZER**

***tran-qui-lax-ant** (tran'kwi-lak'sant) [< L. tranquillus, quiet; L. laxare, to loosen, as the muscles]



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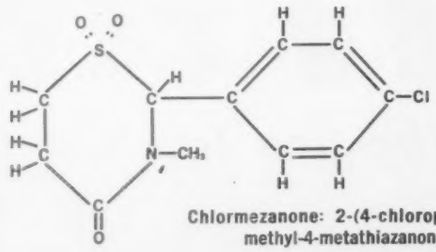
Unrelated chemically to any other drug in current use, Trancopal offers a completely new major chemical contribution to therapeutics.



Chlormezanone: 2-(4-chlorophenyl)-3-methyl-4-metathiazanone-1-dioxide

Clinical studies of over 4400 patients by 105 physicians[1] proved Trancopal remarkably effective in musculoskeletal conditions, anxiety and tension states.

MUSCULOSKELETAL DISORDERS

effective in

93%

of 1570 documented cases of

LOW BACK PAIN

(LUMBAGO, SACROILIAC DISORDERS)

By relieving muscle spasm and pain, Trancopal permits early and active exercise and physical therapy to accomplish maximal benefits for rapid recovery.

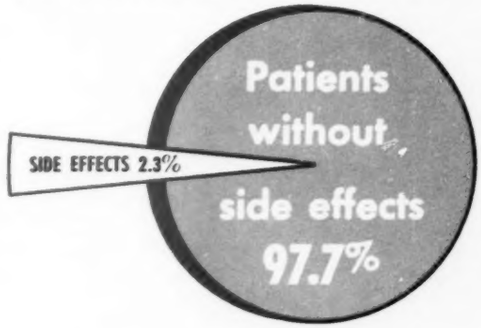
Trancopal

the first true tranquilaxant

BETTER TOLERATED AND SAFER THAN OLDER DRUGS[3]

With Trancopal there is no clouding of consciousness, no euphoria or depression. Even in high dosage, there is no perceptible soporific effect. Because it does not irritate gastric mucosa, it can be taken without regard to mealtimes. Administration does not hamper work—or play. Blood pressure, pulse rate, respiration and digestive processes are unaffected by therapeutic dosage. Toxicity is extremely low. And Trancopal has a lower incidence of side effects than has zoxazolamine, methocarbamol or meprobamate.

INCIDENCE OF SIDE EFFECTS WITH TRANCOPAL IN 4483 PATIENTS



ANXIETY AND TENSION STATES

effective in

88%

of 443 documented cases of

DYSMENORRHEA

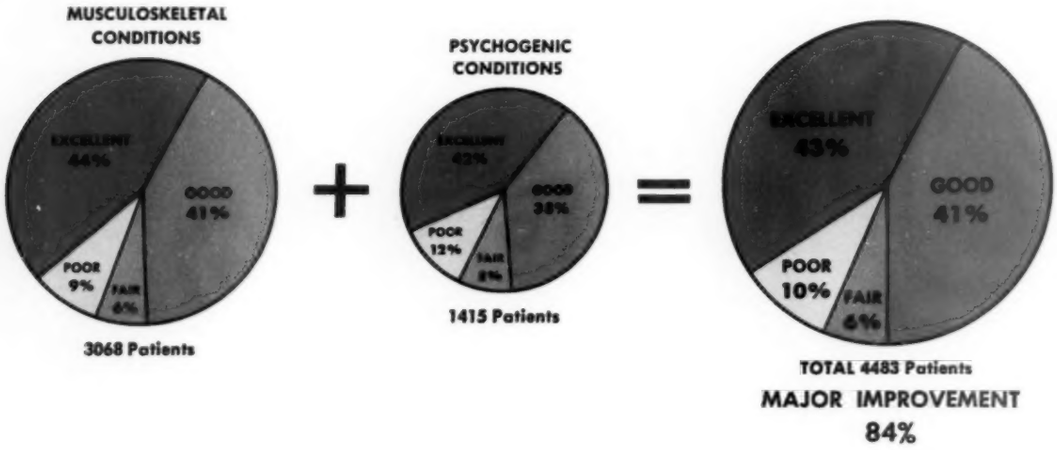
AND PREMENSTRUAL TENSION

Because of its exceptional calmative property, Trancopal "... allows the patient to use his energies in a more productive manner in overcoming his basic problems."[2]

Dosage: 100 to 200 mg. orally three or four times daily. Relief of symptoms occurs in from fifteen to thirty minutes and lasts from four to six hours.

Thoroughly evaluated clinically...

Clinical studies of 4483 patients by 105 physicians[1] have demonstrated that Trancopal often is effective when other drugs have failed. From these studies it is evident that Trancopal can provide more help for a greater number of tense, spastic, and/or emotionally upset patients than can any other chemotherapeutic agent in current use.



INDICATIONS

Musculoskeletal

- Low back pain (lumbago)
- Neck pain (torticollis, etc.)
- Bursitis
- Rheumatoid arthritis
- Osteoarthritis
- Disk syndrome
- Fibrositis
- Ankle sprain, tennis elbow, etc.
- Myositis
- Postoperative muscle spasm

Psychogenic

- Anxiety and tension states
- Dysmenorrhea
- Premenstrual tension
- Asthma
- Angina pectoris
- Alcoholism

Supplied: Trancopal Caplets® (scored) 100 mg., bottles of 100.

References: 1. Collective Study, Department of Medical Research, Winthrop Laboratories. • 2. Ganz, S.E: J. Indiana M. A. In press. • 3. Lichtman, A.L.: Kentucky Acad. Gen. Pract. J. 4:28, Oct., 1958.

the first true tranquilaxant

Trancopal

Potent
MUSCLE RELAXANT
...Equally effective as a
TRANQUILIZER

Winthrop LABORATORIES | **New York 18, New York**

Trancopal (brand of chlormezanone) and Caplets, trademarks reg. U.S. Pat. Off.

Printed in U. S. A. (4191A)

Record of patient with congestive failure treated at a leading Philadelphia hospital. Photos used with permission of the patient.

marked pitting edema (4+) cleared in 4 days with Esidrix

4th day

L.S., 81 years old, admitted to hospital 3/3/59 with hematuria, arteriosclerotic cardiovascular disease, poorly compensated heart failure. When 4+ pitting edema, hepatic congestion and râles failed to clear on regimen of salt restriction, digitalis and pulmonary decongestants by 3/6, Esidrix 50 mg. b.i.d. was ordered. By 3/8 L.S. had lost 3 pounds, pitting edema was reduced to 1+, râles decreased. Patient was ambulatory on 4th day of Esidrix therapy; no evidence of edema. He was discharged feeling well on 3/14.

Now...for edema and hypertension

Dosage: Average daily dosage (orally) is 75 to 100 mg., with a range of 25 to 200 mg. A single dose may be given in the morning or tablets may be administered 2 or 3 times a day.

Supplied: Tablets, 25 mg. (pink, scored); bottles of 100 and 1000. *Tablets,* 50 mg. (yellow, scored); bottles of 100 and 1000.

CIBA
SUMMIT N J

Esidrix T.M.

(hydrochlorothiazide CIBA)

■ relieves edema in many patients refractory to other diuretics ■ often produces greater weight loss than parenteral mercurials or chlorothiazide ■ provides greater average reduction in blood pressure than chlorothiazide ■ is exceptionally safe 2/2688HX

remember **Serpasil®** (reserpine CIBA) | for the anxious hypertensive with or without tachycardia

In HYPERTENSION change to MODERIL® brand of rescinnamine the rewards of reserpine for better management

Gradual, sustained lowering of blood pressure

Effective alone in mild to moderate labile hypertension

Adjunctive therapy in severe hypertension, cutting dosage requirements and side effects of other agents

DOSAGE: Up to 0.5 mg. daily for 2 weeks; then adjust to patient response.

Supplied: Tablets, 0.25 mg. (yellow) and 0.5 mg. (salmon).

"as effective an hypotensive agent as reserpine... producing less side effects"[1]

"a safe, easily tolerated agent for chronic drug administration..."[2]

"produces less sedation and less bradycardia in proportion to its hypotensive dose than does reserpine."[3]

"Reserpine produces more weakness, fatigue, and sedation... In this series reserpine produced mental depression in a number of cases, and this was not observed with rescinnamine [MODERIL]*..."*[4]

"Sedation and bradycardia, in particular, appear to occur less frequently and in mild form with rescinnamine."[5]

"mild depressions, nightmares or excessive drowsiness have been relieved on transfer to equally hypotensive doses of... rescinnamine. The increase of appetite that often follows the administration of reserpine has been lacking and even replaced by decrease of appetite on transfer to rescinnamine."[6]

"rescinnamine has less slowing effect on the heart than does reserpine and apparently has less gastrointestinal effect."[2]

"the addition of rescinnamine reduced the need for high doses of ganglion-blocking agents."[1]

without its penalties

MODERIL—a purified alkaloid of Rauwolfia—lessens the frequency and severity of reserpine side effects,[1-10] *such as:*

sedation • bradycardia •
mental depression • weakness
• fatigue • lassitude •
sleepiness • nightmares
• gastrointestinal effects

MODERIL BIBLIOGRAPHY 1. Winton, S. S.: Internat. Rec. Med. 170:665, 1957. 2. Hershberger, R. L.; Dennis, E. W., and Moyer, J. H.: Am. J. M. Sc. 231:542, 1956. 3. Moyer, J. H.: A. M. A. Arch. Int. Med. 96:518, 1955. 4. Moyer, J. H.; Dennis, E., and Ford, R.: Ibid. p. 530. 5. Council on Drugs, A. M. A.: New and Nonofficial Drugs 1959, Philadelphia, Lippincott, 1959, p. 374. 6. Smirk, F. H.: Postgrad. Med. 25:11, 1959. 7. Moyer, J. H., et al.: South. M. J. 50:499, 1957. 8. Malamud, W., et al.: Am. J. Psychiat. 114:193, 1957. 9. Smirk, F. H., and McQueen, E. G.: Lancet 2:115, 1955. 10. Malitz, S.: J. Chron. Dis. 9:278, 1959.

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Division, Chas. Pfizer & Co., Inc.

NOW

. . . a new way to relieve pain and stiffness in muscles and joints

INDICATED IN:

- MUSCLE STIFFNESS
- LUMBOSACRAL STRAIN
- SACROILIAC STRAIN
- WHIPLASH INJURY
- BURSITIS
- SPRAINS
- TENOSYNOVITIS
- FIBROSITIS
- FIBROMYOSITIS
- LOW BACK PAIN
- DISC SYNDROME
- SPRAINED BACK
- "TIGHT NECK"
- TRAUMATIC STRAINS AND BRUISES
- POSTOPERATIVE MYALGIA

■ Exhibits unusual analgesic properties, different from those of any other drug ■ Specific and superior in relief of SOMAtic pain

■ Modifies central perception of pain without abolishing natural defense reflexes ■ Relaxes abnormal tension of skeletal muscle

SOMA™ T.M.

N-isopropyl-2-methyl-2-propyl-1, 3-propanediol dicarbamate

■ More specific than salicylates ■ Less drastic than steroids

■ More effective than muscle relaxants

SOMA has an unique analgesic action. It apparently modifies central pain perception without abolishing peripheral pain reflexes. SOMA is particularly effective in relieving joint pain. Patients say that they feel better and sleep better with SOMA than with any previously used analgesic, sedative or relaxant drug.
SOMA also relaxes muscle hypertonia, with its stresses on related joints, ligaments and skeletal structures.

ACTS FAST. Pain-relieving and relaxant effects start in 30 minutes and last 6 hours.

NOTABLY SAFE. Toxicity of SOMA is extremely low. No effects on liver, endocrine system, blood pressure, blood picture or urine have been reported. Some patients may become sleepy on high dosage.

EASY TO USE. Usual adult dose is one 350 mg. tablet 3 times daily and at bedtime.

SUPPLIED: Bottles of 50 white sugar-coated 350 mg. tablets.

Literature and samples on request.

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efficacy
economy
rapidity
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adaptability

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Reports in hundreds
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scores of standard textbooks
reflect the position of Gantrisin as drug
of choice in urinary and other bacterial infections.

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control

virtually all *runaway diarrheas..* promptly, effectively with

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Prompt and more dependable control of virtually all diarrheas can be achieved with the comprehensive DONNAGEL formula, which provides adsorbent, demulcent, antispasmodic and sedative effects—with or without an antibiotic. Early re-establishment of normal bowel function is assured—for all ages, in all seasons.

DONNAGEL: In each 30 cc. (1 fl. oz.):

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Pectin (2 gr.)	142.8 mg.
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Atropine sulfate	0.0194 mg.
Hyoscine hydrobromide	0.0065 mg.
Phenobarbital (¼ gr.)	16.2 mg.

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Same formula, plus

Neomycin sulfate	300 mg.

(Equal to neomycin base, 210 mg.)

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Tofrānil®
brand of imipramine HCl

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An entirely new compound originated in the Geigy research laboratories, Tofrānil:

Exercises specific selective action on the symptoms of depressions

Is effective in 70-85% of cases

Is frequently successful in even the most profound and chronic cases

Is virtually devoid of serious side reactions

Reduces the need for electro-convulsive therapy

May be administered by either oral or intramuscular routes

Indications for Tofrānil include:
Endogenous Depression
Reactive Depression
Involutional Melancholia
Senile Depression
Depression Associated with Other Psychiatric Disorders

Detailed literature available on request.

In order to insure optimal results with minimal risk of side reactions physicians are urged to study the "Statement of Directions" before prescribing Tofrānil.

Availability:
Sugar-coated, coral-colored tablets, 25 mg. each, in bottles of 100. Ampuls for intramuscular administration only, each containing 25 mg. in 2 cc. of solution (1.25 per cent) in cartons of 10 and 50.

Selected Bibliography:

1. Ayd, F. J., Jr.: Bulletin of School of Medicine, University of Maryland *44*:29, 1959.
2. Azima, H.: Canad. M.A.J. *80*:535, 1959.
3. Azima, H., and Vispo, R. H.: Am. J. Psychiat. *115*:245, 1958.
4. Kuhn, R.: Am. J. Psychiat. *115*:459, 1958.
5. Lehmann, H. E.; Cahn, C. H., and de Verteuil, R. L.: Canad. Psychiat. A. J. *3*:155, 1958.
6. Mann, A. M., and MacPherson, A. S.: Canad. Psychiat. A. J. *4*:38, 1959.
7. Sloane, R. B.; Habib, A., and Batt, U. E.: Canad. M.A.J. *80*:540, 1959.
8. Straker, M.: Canad. M.A.J. *80*:546, 1959.

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Specific in the treatment of depression the new agent Tofrānil is of unsurpassed potency and effectiveness. Not to be confused with the tranquilizers...Tofrānil ameliorates pathologic depressive states but does not act as a stimulant in the absence of depression. The term "thymoleptic," suggestive of uplifting the spirits, has been chosen to designate this unique type of action.

Tofrānil®
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potent and unique thymoleptic

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IMPROVED

TRICOFURON®

destroys all 3 *principal pathogens*

Whether vaginitis is caused by Trichomonas, Monilia or Hemophilus vaginalis—alone or combined—TRICOFURON IMPROVED swiftly relieves symptoms and malodor, and achieves a truly high percentage of cultural cures, frequently in 1 menstrual cycle. TRICOFURON IMPROVED provides: a *new* specific moniliacide MICOFUR® brand of nifuroxime, the *established* specific trichomonacide FUROXONE® brand of furazolidone and the *combined* actions of both against Hemophilus vaginalis.

1. *Office* insufflation once weekly of the Powder (MICOFUR [*anti*-5-nitro-2-furaldoxime] 0.5% and FUROXONE 0.1% in an acidic water-soluble powder base). **2.** Continued *home* use twice daily, with the Suppositories (MICOFUR 0.375% and FUROXONE 0.25% in a water-miscible base).

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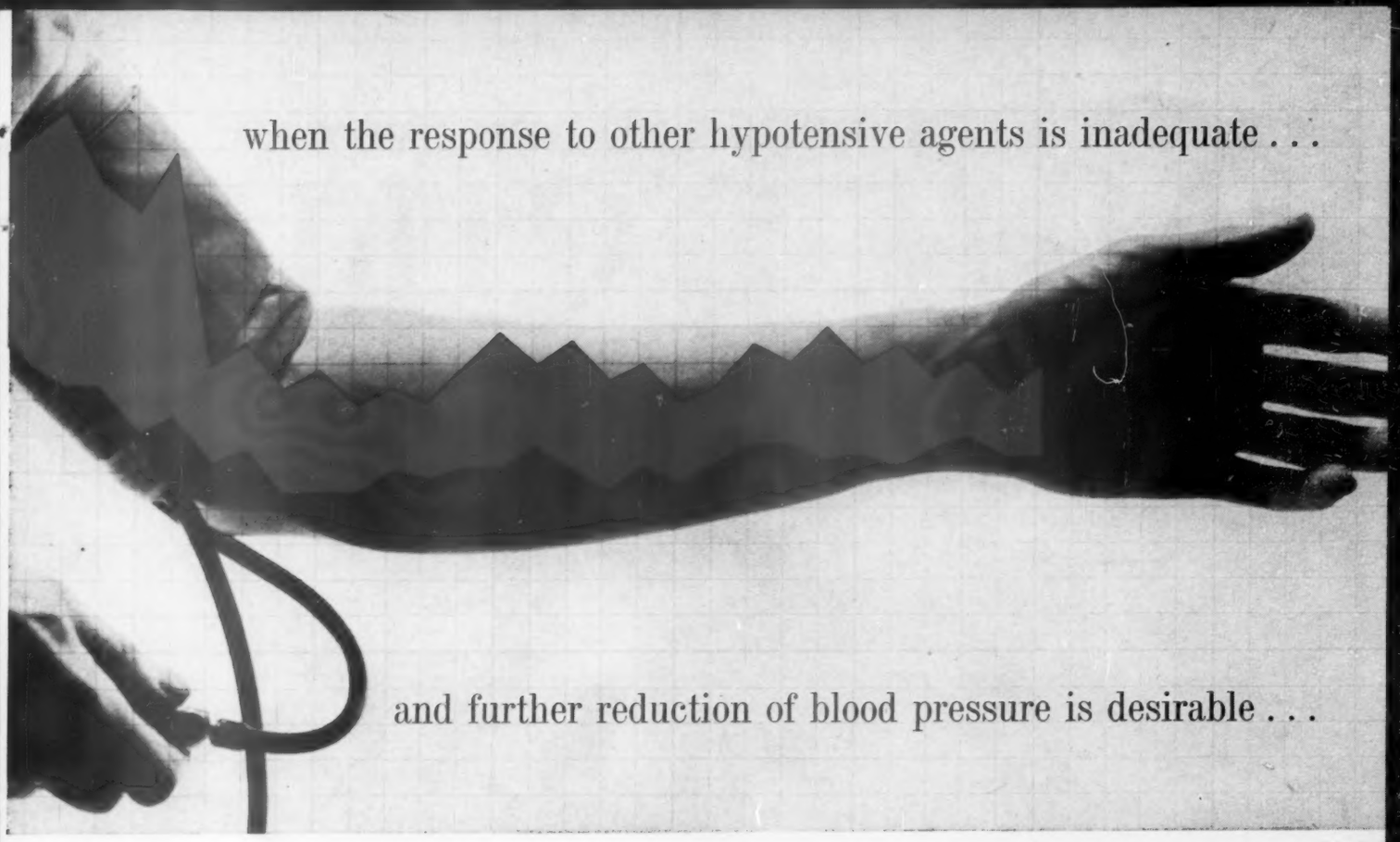

adding ganglionic-blocking

INVERSINE®

MECAMYLAMINE HCl

often makes possible a lessening of cardiovascular-renal damage, regression of the basic disease, and prolongation of life

"unnecessary delay must be avoided in establishing ganglion blockade in severe or malignant hypertension"
Beem, J. R., and Moyer, J. H.: Geriatrics 13:378, June 1958.

relieve high blood pressure and its manifestations

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for moderate, severe, and malignant hypertension

"When employed under carefully controlled conditions with adequate attention to proper regulation of dosage, mecamylamine ['INVERSINE'] may be expected to reduce blood pressure effectively and to ameliorate various manifestations of hypertensive-cardiovascular disease. These include such symptoms as headache, dizziness, vertigo, hypertensive encephalopathy, cerebral or subarachnoid hemorrhage, retinopathy, cardiac hypertrophy and, in some cases, cardiac decompensation."

A.M.A. Council on Drugs, New and Nonofficial Drugs: Philadelphia, J. B. Lippincott Co., 1958, p. 285

A GREATLY IMPROVED GANGLIONIC BLOCKING AGENT

'INVERSINE'

- of the orally effective blocking agents, only 'INVERSINE' is completely and uniformly absorbed
- because it is uniformly absorbed, 'INVERSINE' provides predictable, reproducible effects with minimal day-to-day fluctuations
- has a gradual onset of effect, reducing the likelihood of sudden drops in blood pressure
- effective in extremely low dosage (orally, 10 mg. 'INVERSINE' is approximately equivalent to 100 mg. pentolinium, 80 mg. chlorisondamine, 1000 mg. hexamethonium)
- has a long duration of action (6 to 12 hours or longer), permitting convenient dosage schedules
- development of tolerance is not as pronounced as with other ganglionic blocking drugs
- effective in many patients who do not respond to other ganglionic blocking drugs

pretreatment with 'Diuril', or 'Diuril' and rauwolfia, enhances therapy with 'Inversine'

"Pretreatment with chlorothiazide ['DIURIL'] and rauwolfia reduces the dosage requirement, augments blood pressure response, and moderates certain of the side effects of ganglion blocking agents. Although such basal therapy is advantageous, unnecessary delay must be avoided in establishing ganglion blockade in severe or malignant hypertension."

Beem, J. R., and Moyer, J. H.: Geriatrics 13:378, June 1958

dosage recommendations for new patients

1. *Initiate therapy with 'DIURIL'*

'DIURIL' is given in a dosage range of from 250 mg. twice a day to 500 mg. three times a day, depending on severity of the hypertension.

2. *Add other agents*

Other drugs (rauwolfia, 'INVERSINE', hydralazine, etc.) are added as necessary and their dosage adjusted according to patient response.

'INVERSINE' is given in the same manner whether used with other drugs or alone. Recommended initial dosage is 2.5 mg. twice a day, preferably after meals. May be increased by 2.5 mg. at intervals of no less than two days until desired response is obtained. In severe or urgent cases, the increments may have to be larger or more frequent, with the largest dose given preferably at noon or in the evening. 'INVERSINE' is extremely potent and should always be titrated according to the patient's orthostatic blood pressure response.

3. *Adjust dosage of all medication*

The patient must be observed frequently and careful adjustment of all agents should be made to determine optimal maintenance dosage.

Precautions: Side effects of 'INVERSINE' are essentially the same as those encountered with other ganglionic blocking agents. At the first sign of constipation, vigorous treatment must be initiated immediately since paralytic ileus may result if constipation is unchecked. Patients should be informed how to cope with postural hypotension should this occur. 'INVERSINE' is contraindicated in coronary insufficiency, organic pyloric stenosis and recent myocardial infarction. Additional information on 'INVERSINE' and 'DIURIL' is available on request.

Supplied: 'INVERSINE', tablets of 2.5 and 10 mg. Bottles of 100.

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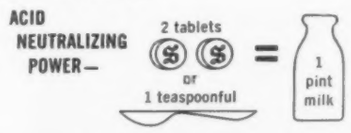

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in the depressed, unhappy patient

PROMPTLY IMPROVES MOOD

without excitation

- **Acts fast to relieve depression and its common symptoms:** sadness, crying, anorexia, listlessness, irritability, rumination, and insomnia.
- **Restores normal sleep**—without hang-over or depressive aftereffects. Usually eliminates need for sedative-hypnotics.

EFFICACY AND SAFETY CONFIRMED IN OVER 3,000 DOCUMENTED CASE HISTORIES.[1, 2, 3]

Dosage: Usual starting dose is 1 tablet q.i.d. When necessary, this dose may be gradually increased up to 3 tablets q.i.d.

Composition: Each light-pink, scored tablet contains 1 mg. 2-diethylaminoethyl benzilate hydrochloride (benactyzine HCl) and 400 mg. meprobamate.

References:

1. Alexander, L.: J.A.M.A. 166:1019, March 1, 1958.
2. Current personal communications; in the files of Wallace Laboratories.
3. Pennington, V.M.: Am. J. Psychiat. 115:250, Sept. 1958.

for depression

▲Deprol▲†

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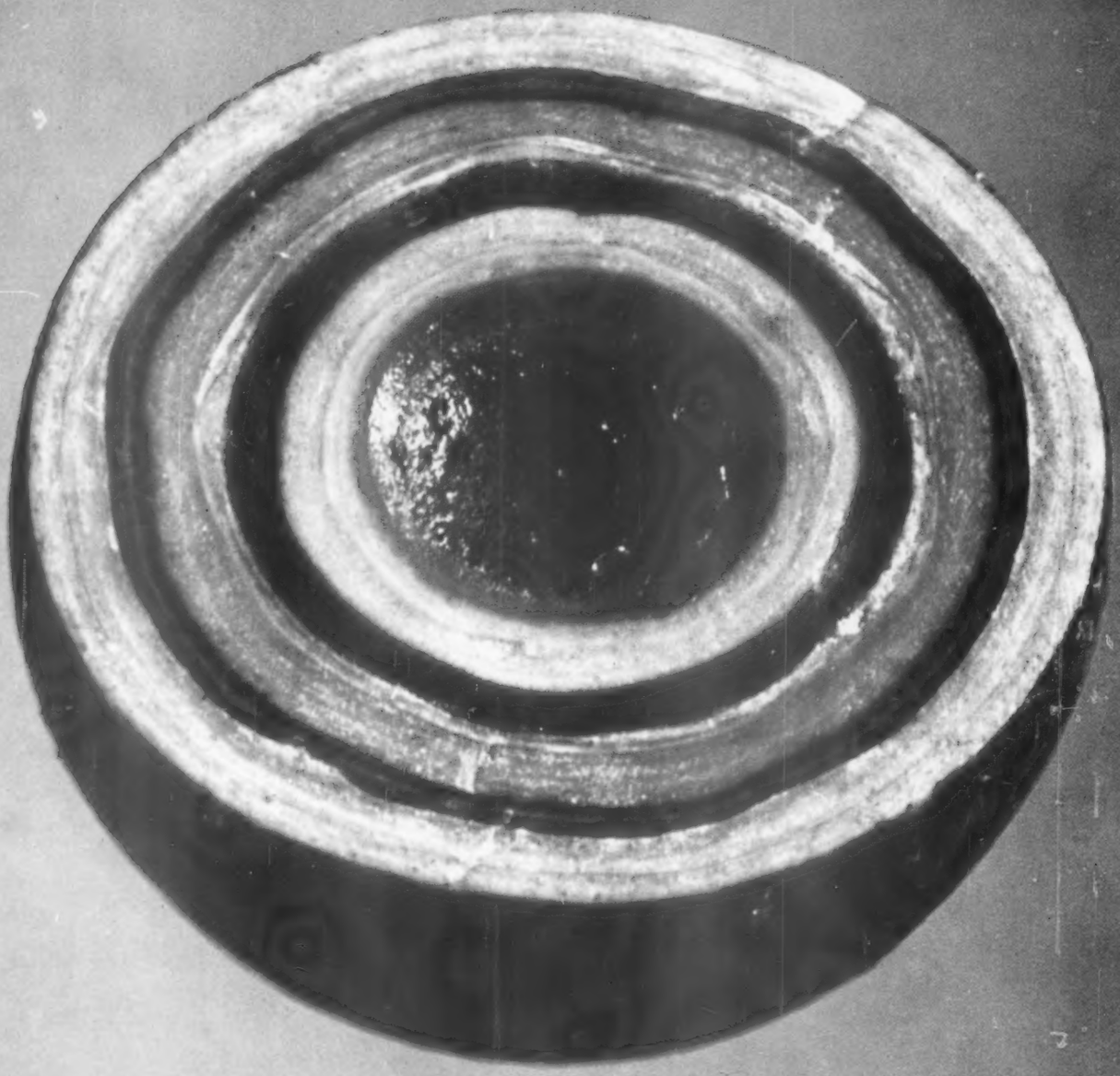

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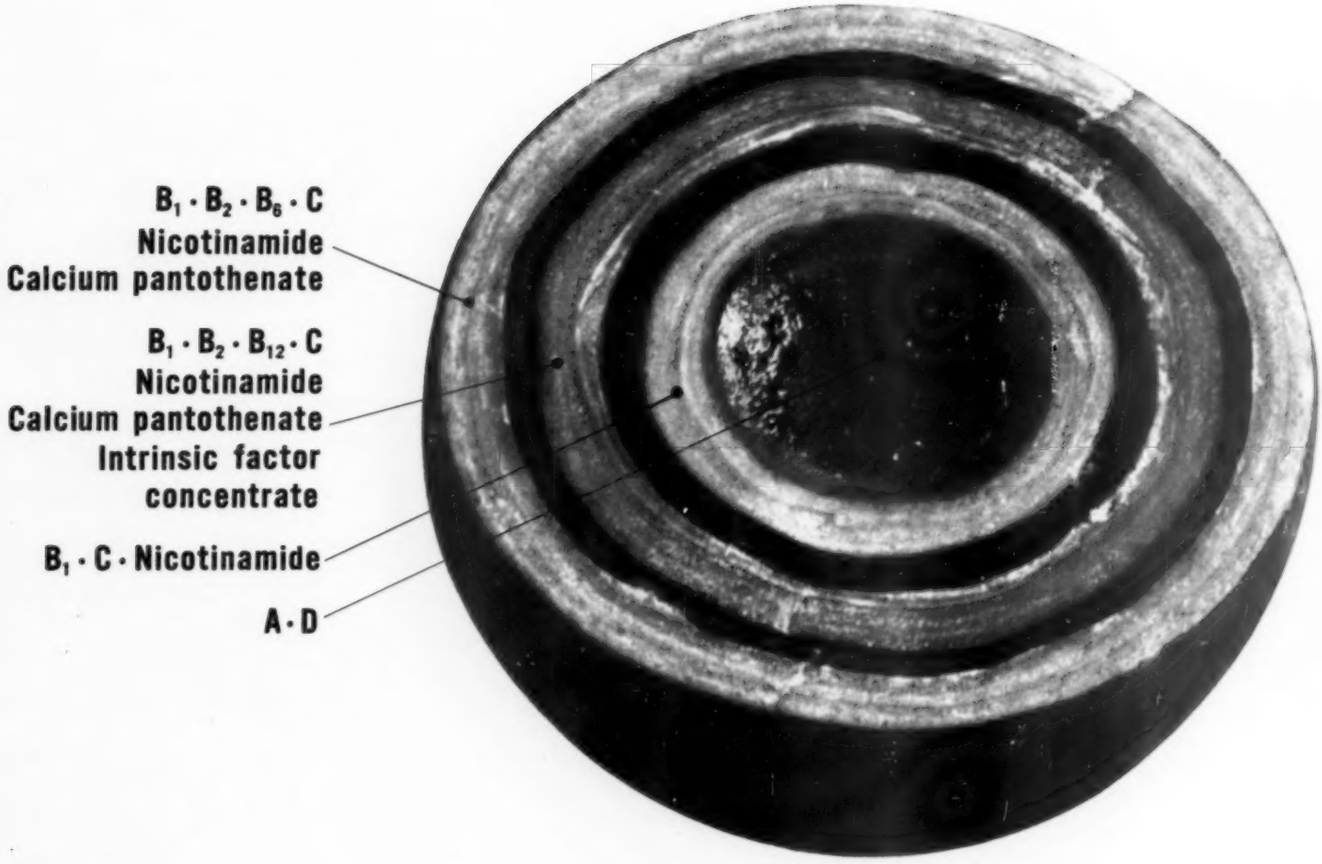

SEQUENCE OF CONTROLLED RELEASE*

Each tablet contains:	1. The outer layer dissolves, releasing:	2. The middle layer releases:	3. The core releases:
Vitamin A .... 25,000 U.S.P. Units			25,000 U.S.P. Units
Vitamin D ..... 1,000 U.S.P. Units			1,000 U.S.P. Units
Vitamin B_1 mononitrate.. 25.0 mg.	10.0 mg.	10.0 mg.	5.0 mg.
Vitamin B_2 ..... 12.5 mg.	7.5 mg.	5.0 mg.	
Nicotinamide ..... 50.0 mg.	20.0 mg.	20.0 mg.	10.0 mg.
Vitamin B_6 ..... 3.0 mg.	3.0 mg.		
Calc. pantothenate ..... 10.0 mg.	5.0 mg.	5.0 mg.	
Vitamin C .....250.0 mg.	100.0 mg.	100.0 mg.	50.0 mg.
Vitamin B_{12} ..... 5.0 mcg.		5.0 mcg.	
Intrinsic factor concentrate 15.0 mg.		15.0 mg.	
*Timetable of controlled release *in vitro*	½—¾ hr.	1½ hrs.	3 hrs.

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Now, for the first time, a multivitamin tablet offers the benefits of *divided dosage with a single administration.*

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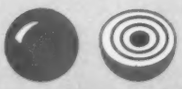

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Dosage: One tablet daily, preferably at mealtime.

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5936

A new menu book for your patients with serum cholesterol problems

saves your time because it answers patients' questions... helps them follow your diet instructions.

We have just published "Cooking For Health With Mazola Corn Oil," to help you with patients who need to increase the proportion of unsaturated fats in their diets.

It contains answers to literally dozens of questions your patients will ask on the subject of cholesterol and diet.

It will be a helpful guide in planning meals which provide desirable proportions of linoleate-rich unsaturated fats.

It contains many tempting recipes which are low in saturated fat.

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For your copy of "Cooking For Health With Mazola Corn Oil" and a supply of postage-free cards by which your patients may order their copies, simply mail us the coupon below.

NEW CONCEPTS OF THE VALUE OF CORN OIL TO HEALTH

Clinical studies with controlled diets have demonstrated that corn oil can be effective in lowering serum cholesterol levels. Investigators have variously ascribed this action of corn oil to total unsaturation, linoleic acid, sitosterols and other active components not as yet identified.

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and other abnormal levels of serum gamma globulin

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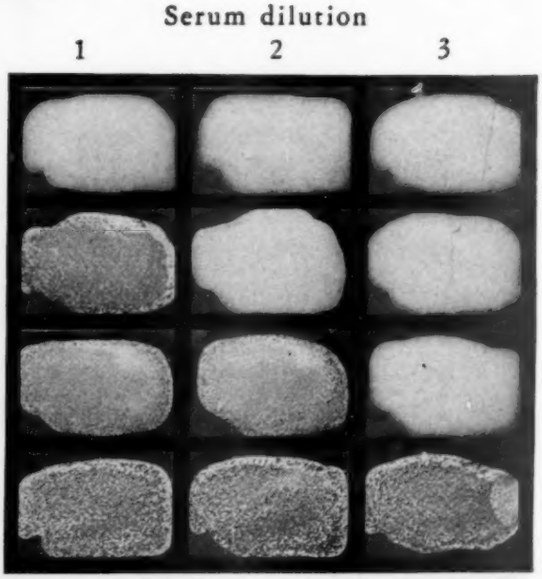

*Trade Mark of Hyland Laboratories

Hyland GG-TEST requires only one drop of patient's serum and is performed simply and quickly on a glass slide (reactions in less than 2 minutes). Permits routine GG serum level determinations at low cost without overtaxing laboratory facilities.

Supplied: Kit containing Latex-Anti-Human Gamma Globulin Reagent and Glycine-Saline Buffer Diluent for 40 tests, Normal Serum Controls diluted ready for use, capillary pipettes for preparing serum dilutions, divided glass slide, and complete directions for the test. $15.00 per kit.

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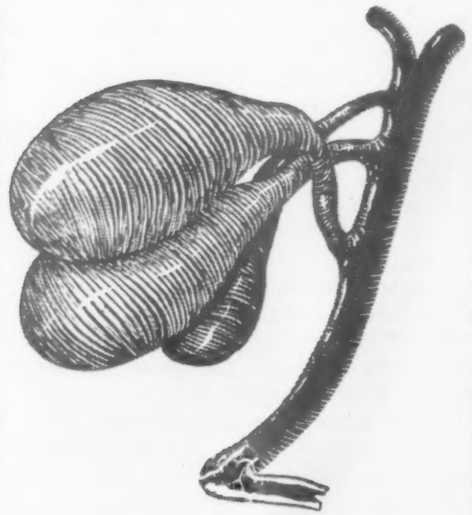
AN AMES CLINIQUICK™

CLINICAL BRIEFS FOR MODERN PRACTICE

HOW PREVALENT ARE MULTIPLE GALLBLADDER ANOMALIES?

One hundred and twenty-two cases of *vesica fellea divisa* (bilobed gallbladder) and *vesica fellea duplex* (double gallbladder with 2 cystic ducts) are reported in the literature. A unique case of *vesica fellea triplex* has recently been described.

Source: Skilboe, B.: Am. J. Clin. Path. *30*:252, 1958.



in medical management and postoperative care of biliary disorders...

"effective" hydrocholeresis...

DECHOLIN®

(dehydrocholic acid, AMES)

"...dehydrocholic acid...does considerably increase the volume output of a bile of relatively high water content and low viscosity. This drug is therefore a good 'flusher,' and is effectively used in treating both the chronic unoperated patient and the patient who has a T-tube drainage of an infected common bile duct."[1]

free-flowing bile
plus reliable spasmolysis

DECHOLIN® WITH BELLADONNA

"...DECHOLIN/Belladonna in a dosage of one tablet t.i.d. for a period of two to three months may prove helpful in relieving postoperative symptoms, aiding the digestion, and facilitating elimination."[2]

(1) Beckman, H.: Drugs: Their Nature, Action and Use, Philadelphia, W. B. Saunders Company, 1958, p. 425.
(2) Biliary Tract Diseases, M. Times *85*:1081, 1957.

AMES COMPANY, INC
Elkhart • Indiana
Toronto • Canada

64659

NEW
A SENTRY FOR THE G.I. TRACT

protects against
hypersecretion • hypermotility
hyperirritability • hyperemotivity

ENARAX

anticholinergic / antispasmodic / tranquilizer

A remarkably long-acting anticholinergic. Only one 10 mg. dose of new long-acting oxyphencyclimine controls hypersecretion and spasm[3] for 12 hours or more. In the most recent study at Cook County Hospital, investigators were impressed with its antisecretory effect, leading to prolonged periods of achlorhydria.[6] 51 out of 57 patients with various G.I. disorders were relieved of symptoms on only 2 daily doses.

Plus ATARAX—the antisecretory tranquilizer. Not only does ATARAX modify tension—its added antisecretory action[4,7-9] augments the efficacy of oxyphencyclimine. The combination, ENARAX, freed 100 out of 103 patients of G.I. symptoms.[2] Improvement was especially notable in cases of peptic ulcer, where the emotional factor figures so prominently.

"Side reactions were uncommon...."[2] Selective postganglionic action on the G.I. tract minimizes side effects. Mouth dryness—the most common reaction—seldom reaches troublesome proportions.

Each ENARAX tablet contains: Oxyphencyclimine HCl, 10 mg.; Hydroxyzine HCl (ATARAX®), 25 mg.

Dosage: One-half to one tablet twice daily—preferably in the morning and before retiring. The maintenance dose should be adjusted according to therapeutic response. Use with caution in patients with prostatic hypertrophy or glaucoma.

Supplied: In bottles of 60 black-and-white scored tablets.

SUMMARY OF CASES

Clinical Diagnosis	*Oxyphencyclimine[1,3,5,6]	†ENARAX[2,5]
Peptic ulcer	440	48
Gastritis	16	17
Gastroenteritis	—	55
Colitis	37	—
Duodenitis	6	3
Functional bowel syndrome	14	—
Hiatus hernia (symptomatic)	16	1
Pylorospasm or cardiospasm	11	2
Irritable bowel	11	—
Biliary tract dysfunctions	11	1
Miscellaneous	7	29
Total number of patients	569	156
Clinical Results *Excellent*	445	150
Fair	56	—
Failure	68	6

* **Oxyphencyclimine alone**—clinically effective in 87% after a year's testing.

† **ENARAX** (oxyphencyclimine plus ATARAX)—all successful cases in "excellent" category.

RAX®

A SENTRY FOR THE G.I. TRACT

one tablet at breakfast
one tablet at bedtime
for *full-time* relief

(oxyphencyclimine plus ATARAX®)

ACID REDUCTION AFTER OXYPHENCYCLIMINE THERAPY

Tests conducted in 9 representative ulcer patients after overnight fasts showed considerable reduction in both volume and acidity.

References: 1. Winkelstein, A.: Am. J. Gastroenterol., in press. 2. Leming, B. H., Jr.: Clin. Med. *6*:423 (March) 1959. 3. McHardy, G., et al.: Paper presented at Postgraduate Course in Gastroenterology, University of California School of Medicine, San Francisco, Calif., January 27, 1958. 4. Strub, I. H., and Carballo, A.: To be published. 5. Data in Roerig Medical Department files. 6. Steigmann, F.: To be published. 7. Schuller, E.: Gaz. des Hôpitaux *10*:391 (Apr. 10) 1957. 8. Farah, L.: Internat. Rec. Med. *169*:379 (June) 1956. 9. Harrisson, J. W. E., et al.: Paper presented at the 4th Pan-American Congress of Pharmacy and Biochemistry, Washington, D. C., November 3-9, 1957.

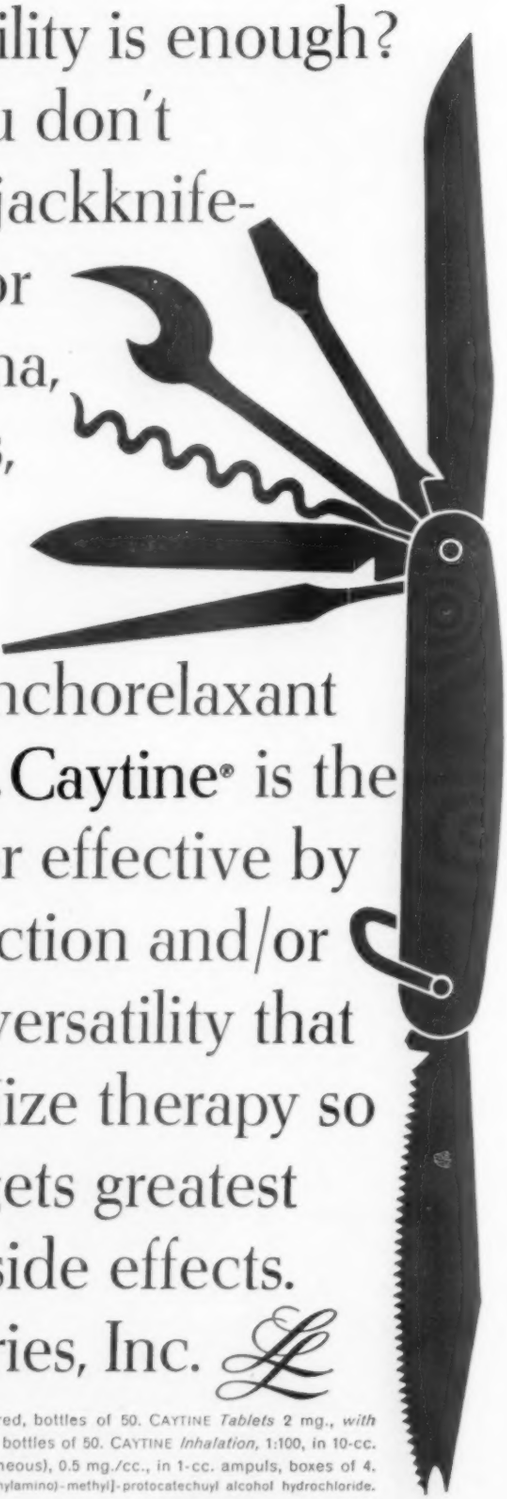
ROERIG

New York 17, N. Y.
Division, Chas. Pfizer & Co., Inc.
Science for the World's Well-Being

How much versatility is enough?

That depends. You don't ordinarily need a jackknife-of-*all*-trades. But for asthma, emphysema, chronic bronchitis, bronchiectasis, you do need the most versatile bronchorelaxant you can prescribe. **Caytine®** is the first bronchodilator effective by mouth and/or injection and/or inhalation. Here's versatility that lets you individualize therapy so that each patient gets greatest relief with fewest side effects.

Lakeside Laboratories, Inc.



CAYTINE—*3 effective forms:* CAYTINE *Tablets Plain,* 2 mg., scored, bottles of 50. CAYTINE *Tablets* 2 mg., *with Pentobarbital,* 32 mg. (warning: may be habit forming), scored, bottles of 50. CAYTINE *Inhalation,* 1:100, in 10-cc. bottles with dropper. CAYTINE *Injection* (intramuscular, subcutaneous), 0.5 mg./cc., in 1-cc. ampuls, boxes of 4. CAYTINE is the only brand of the α[(α-methyl-3,4-methylenedioxyphenethylamino)-methyl]-protocatechuyl alcohol hydrochloride.

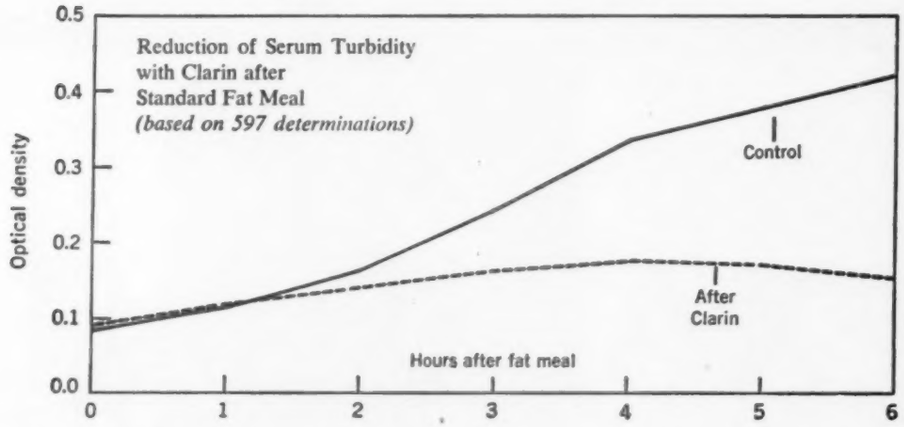
48559

in the management of atherosclerosis

Clarin*

(sublingual heparin potassium, Leeming)

clears lipemic serum



Each time your patients eat a substantial fat-containing meal, lipemia results. Small amounts of injected heparin will help control this increased fat content in the blood,[1,2] but widespread adoption of this method has been hampered by its inconvenience, pain, cost and the necessity for periodic checks on blood clotting time.

Now, long-term preventive heparin therapy is practical for the first time with the introduction of CLARIN—which is heparin in *sublingual* form. Each CLARIN tablet contains 1500 I.U. of heparin potassium—a sufficient amount to clear lipemic serum without affecting coagulation mechanisms.[3,4]

With one mint-flavored CLARIN tablet under the tongue after each meal, lipemia is regularly controlled, removing a constant source of danger to the atherosclerotic patient. He may eat safely, with less fear of dangerous results, without hard-to-follow diets.

The varied implications of CLARIN in beneficially affecting fat metabolism are obviously far-reaching. The relationship between heparin, lipid metabolism and atherosclerosis may well be analogous to that between insulin, carbohydrate metabolism and diabetes mellitus.[5]

Use CLARIN to protect your atherosclerotic patients—the postcoronaries and those with early signs of coronary artery disease.

Indication: For the management of hyperlipemia associated with atherosclerosis.

Dosage: After each meal, hold one tablet under the tongue until dissolved.

Supplied: In bottles of 50 pink, sublingual tablets, each containing 1500 I.U. heparin potassium.

1. Council on Drugs, J.A.M.A. *166*:52 (Jan. 4) 1958. 2. Hahn, P. F.: Science *98*:19 (July 2) 1943. 3. Fuller, H. L.: Angiology *9*:311 (Oct.) 1958. 4. Rubio, F. A., Jr.: Personal communication. 5. Engelberg, H., *et al.*: Circulation *13*:489 (April) 1956.

*Trade Mark. Patent applied for.

Thos. Leeming & Co., Inc.

155 East 44th Street, New York 17, N. Y.

High-Speed . . . . . DEPENDABLE PRESSURE STEAM STERILIZATION

AMERICAN STERILIZER
613-R PORTABLE DYNACLAVE

• The low cost, high performing American 613-R Dynaclave assures positive sterilization with pressure steam at 250°F. or 270°F. It is fast, reaching 270° F. in approximately seven minutes.

Operation is fully automatic with selective sterilizing cycles from 3 to 60 minutes. Cools and dries instruments or supplies by exhausting steam and residual water back into water reservoir—NOT into room.

The 613-R, with greater capacity, accommodates three large trays (6" x 13"). Handsome, all stainless steel construction is durable and easy to clean. Other features include a Safety-Lock Door, Adjustable Thermostat and Accurate Temperature Gauge. Automatically burn-out proof.

See your authorized American Sterilizer dealer or write for Bulletin DC-410.

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SQUIBB ANNOUNCES NEW

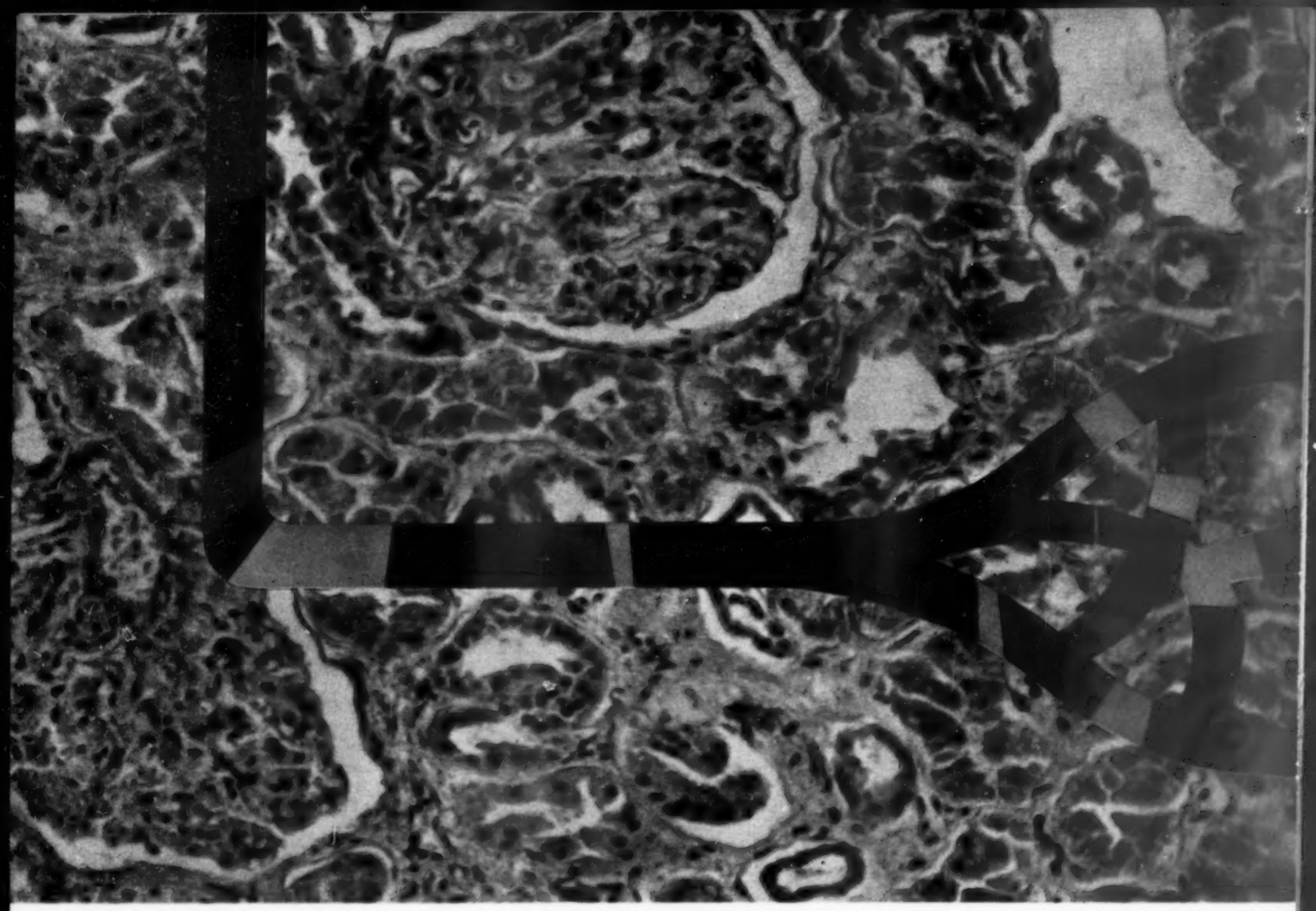
RAUTRAX

a logical combination — Raudixin enhanced by an entirely new diuretic... Flumethiazide

SQUIBB

thus Squibb offers you greater latitude in solving the problem of

HYPERTENSION



RAUT

for the treatment of hypertension...without

Rautrax combines Raudixin with flumethiazide for control of all degrees of hypertension. Clinicians report it safely and rapidly eliminates excess extracellular sodium and water without potassium depletion.[1,2,3] Through this dependable diuretic action of flumethiazide, the clinical and subclinical edema—so often associated with cardiovascular disease—is rapidly brought under control.[2,3,4,5] Flumethiazide also potentiates the antihypertensive action of Raudixin. By this unique dual action, a lower dosage of each ingredient effectively maintains safe antihypertensive therapy.

Diuresis without serum electrolyte imbalance

Flumethiazide—*the new, safe nonmercurial diuretic*—rapidly achieves its diuretic effect without causing appreciable plasma electrolyte imbalance.[1,2,3] Potassium loss is less than with other nonmercurial diuretics.[1] Moreover, the inclusion of supplemental potassium chloride in Rautrax provides added protection against potassium

RAX

RAUDIXIN
SQUIBB STANDARDIZED
WHOLE ROOT RAUWOLFIA SERPENTINA

FLUMETHIAZIDE

POTASSIUM CHLORIDE

fear of significant potassium depletion[1, 2, 3]

and chloride depletion in the long-term management of the hypertensive patient.

Sodium and water retention is rapidly relieved,[2] and once the fluid balance has been brought within normal limits, continued administration of Rautrax does not appreciably alter the normal serum electrolyte pattern.

Control of hypertension with fewer side effects
Raudixin—a cornerstone on which to build a therapeutic regimen for control of hypertension.

RAUTRAX . . . GREATER LATITUDE IN SOLVING THE PROBLEM OF HYPERTENSION

- **Prompt, safe antihypertensive effect — by the complementary action of Raudixin and flumethiazide**
- **Less potassium loss than with other nonmercurial diuretics[1]**
- **No loss in effectiveness after continued administration**
- **No influence on blood urea nitrogen, blood count or other hematologic values[5]**
- **Fewer and less severe side effects[6,7]**
- **Less need for severe restriction in sodium intake**
- **Gout, purpura, or allergic reactions not reported**

SQUIBB

RAUDIXIN PLUS AN ENTIRELY NEW DIURETIC...FLUMETHIAZIDE

A NATURAL COMPANION

TO FAMOUS RAUDIXIN TO HELP SOLVE THE PROBLEM...

HYPERTENSION

Dosage: 2 to 6 tablets daily in divided doses initially; may be adjusted within range of 1 to 6 tablets daily in divided doses. Note: In hypertensive patients already on ganglionic blocking agents, veratrum and/or hydralazine, the addition of Rautrax necessitates an immediate dosage reduction of these agents by at least 50%. A similar reduction is also necessary when these ganglionic blocking agents are added to the Rautrax regimen.
Literature available on request.

Supply: Capsule-shaped tablets each providing 50 mg. Raudixin, 400 mg. flumethiazide, and 400 mg. potassium chloride, bottles of 100.

References: 1. Moyer, J.H., and others: Am. J. Cardiol., *3*:113 (Jan.) 1959. • 2. Bodi, T., and others: To be published, Am. J. Cardiol., (April) 1959. • 3. Fuchs, M., and others: Monographs on Therapy, *4*:43 (April) 1959. • 4. Montero, A.C.; Rochelle, J.B., III, and Ford, R.V.: To be published. • 5. Rochelle J.B., III; Montero, A.C., and Ford, R.V.: To be published. • 6. Montero, A.C.; Rochelle, J.B., III, and Ford, R.V.: To be published. • 7. Doffermyer, L.R.; Byrd, C.W., and Lilly, W.H.: North Carolina M.J. *19*:430 (Oct.) 1958.

SQUIBB

Squibb Quality — the Priceless Ingredient

'RAUDIXIN' AND 'RAUTRAX' ARE SQUIBB TRADEMARKS

NOW... AROUND-THE-CLOCK CONTROL OF APPETITE

NEW PRELUDIN®

ENDURETS™

A PROLONGED-ACTION DOSAGE FORM

Clinical experience has long established PRELUDIN as an antiobesity agent distinguished by its efficacy and its relative freedom from undesirable side actions. Now, convenience is added to reliability in ENDURETS... a specially devised long-acting pharmaceutical form.

Just one PRELUDIN ENDURET (75 mg.) tablet after breakfast curbs appetite throughout the day, in the vast majority of cases.

PRELUDIN® (brand of phenmetrazine hydrochloride) ENDURETS™.
Each ENDURET prolonged-action tablet contains 75 mg. of active principle.
PRELUDIN® is also available as scored, square, pink tablets of 25 mg. for 2 to 3 times daily administration.
Under license from C. H. Boehringer Sohn, Ingelheim.

ENDURETS is a Geigy trademark.

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in India, it's called 'Delhi belly'

diarrhea by any name

GASTROENTERITIS
BACILLARY DYSENTERY
PARADYSENTERY
SALMONELLOSIS
DIARRHEA OF THE NEWBORN
NONSPECIFIC DIARRHEA
"SUMMER COMPLAINT"

usually responds rapidly to

Cremomycin®

NEOMYCIN-SULFASUXIDINE®-KAOLIN-PECTIN SUSPENSION

for *rapid* relief of virtually all diarrheas

fruit-flavored, readily accepted by patients of all ages*

Neomycin — rapidly bactericidal against most intestinal pathogens, but is relatively ineffective against such diarrhea-causing organisms as Shigella.

SULFASUXIDINE® — an ideal adjunct to neomycin because it is highly effective against Shigella and certain other neomycin-resistant organisms.

Kaolin and Pectin — coat and soothe the inflamed mucosa, adsorb toxins, help reduce intestinal hypermotility, help provide rapid symptomatic relief.

***For infants, CREMOMYCIN may be administered in the regular bottle feeding since its fine particles easily pass through a standard nursing nipple.**

MSD MERCK SHARP & DOHME, DIVISION OF MERCK & CO., INC., PHILA. 1, PA.

CREMOMYCIN AND SULFASUXIDINE (SUCCINYLSULFATHIAZOLE) ARE TRADEMARKS OF MERCK & CO., INC.

THE HEART DISEASE PATIENT NEEDS RELIEF FROM EMOTIONAL STRESS

ANXIETY INTENSIFIES the physical disorder in heart disease. "The prognosis depends largely on the ability of the physician to control the anxiety factor, as well as the somatic disease."

(Friedlander, H. S.: The role of ataraxics in cardiology. Am. J. Cardiol. 1:395, March 1958.)

TRANQUILIZATION WITH MILTOWN enhances recovery from acute cardiac episodes and makes patients more amenable to necessary limitations of activities.

(Waldman, S. and Pelner, L.: Management of anxiety associated with heart disease. Am. Pract. & Digest Treat. 8:1075, July 1957.)

Miltown®

meprobamate (Wallace)

Available in 400 mg. scored and 200 mg. sugar-coated tablets. Also available as MEPROSPAN* (200 mg. meprobamate *continuous release* capsules). In combination with a nitrate, for angina pectoris: MILTRATE*—(Miltown 200 mg. + PETN 10 mg.).

*TRADE-MARK CM-7726

Miltown causes no adverse effects on heart rate, blood pressure, respiration or other autonomic functions.

WALLACE LABORATORIES, *New Brunswick, N. J.*

for full corticosteroid benefits: new Gammacorten

this arthritic needed Gammacorten

PATIENT M. S., age 81, at time of first visit was in severe pain and very uncomfortable. Complained of swelling of wrists, legs, various joints; there was pain and stiffness in cervical area and lower spine; pain, swelling and limited motion in the fingers; slight ulnar deviation of the hand. He could not raise his arms above the level of his shoulders.

Treatment and Result: After 36 hours of **GAMMACORTEN** therapy, M. S. had "complete relief." Joint swelling had decreased, pain was almost absent, range of motion had increased dramatically. At the end of the first week of **GAMMACORTEN** he was free of discomfort and able to return to his job as a porter.

BEFORE GAMMACORTEN: **M. S. demonstrates the position necessary to put on his hat (range of motion was so restricted that he could not comb his hair).**

AFTER ONE WEEK OF GAMMACORTEN: **M. S. could put on his hat normally, could comb hair; function near-normal at end of first week of treatment.**

BEFORE GAMMACORTEN: **His fingers were extremely painful and were so swollen that a size 11 jeweler's ring would not fit over his small finger.**

AFTER ONE WEEK OF GAMMACORTEN: **Size 11 jeweler's ring passes easily over previously swollen joint. At end of first week, "puffiness" had virtually disappeared.**

BEFORE GAMMACORTEN: **Hands were so painful, stiff and swollen that M. S. could not flatten hand or extend fingers on flat surface.**

AFTER ONE WEEK OF GAMMACORTEN: **Pain completely subsided. M. S. can flatten hand, extend fingers and flex in normal manner without pain.**

Photographs used with permission of patient.

How to use Gammacorten

With **GAMMACORTEN,** a full measure of corticosteroid benefit can now be brought to patients who have heretofore obtained less than optimal benefit from adrenocorticoid therapy. In practice, the increased activity of **GAMMACORTEN** means maximal mobility for the arthritic; maximal freedom from attack for the asthmatic; rapid and complete resolution of lesions for the dermatologic patient. Unwanted adrenocorticoid effects are relatively infrequent with **GAMMACORTEN.** Should side effects occur, they can be usually managed by reducing dosage or by supplemental measures.

in arthritis — An initial dosage of 1.5 to 3 mg. per day (2 to 4 tablets divided into 3 or 4 doses). This dosage should be continued until a satisfactory symptomatic response is obtained — usually within 3 or 4 days. After a favorable response has been obtained, reduce dosage by 1/3 every 2 to 3 days until either maintenance dosage is established or therapy can be discontinued. Satisfactory control can often be maintained with as little as 0.75 mg. to 1.5 mg. per day.

in asthma and allergy — **IN STATUS ASTHMATICUS:** Initial daily dosage of GAMMACORTEN is 7.5 to 10 mg. (10 to 13 tablets divided into 3 or 4 doses). As soon as the acute state is controlled, reduce dosage slowly by 1/3 to 1/4 until a satisfactory maintenance level is reached or until therapy is discontinued.

IN CHRONIC BRONCHIAL ASTHMA: Initial dosage is 1.5 to 3 mg. of GAMMACORTEN per day (2 to 4 tablets divided into 3 or 4 doses). After a satisfactory response has been obtained, decrease dosage by 1/3 every 2 to 3 days until either maintenance level has been determined or therapy can be discontinued. Asthmatics can often be maintained for long periods on as little as 0.75 mg. to 1.5 mg. of GAMMACORTEN daily.

IN INTRACTABLE HAY FEVER: Start with 2 to 3 mg. (3 to 4 tablets divided into 3 or 4 doses) of GAMMACORTEN per day. Symptoms should be promptly relieved; prolonged maintenance therapy is unnecessary for these self-limiting disorders.

in skin disorders Start with 2 to 3 mg. (3 to 4 tablets divided into 3 or 4 doses) of GAMMACORTEN daily. Satisfactory control is usually obtained at this dosage level. In chronic conditions, dosage should be decreased by 1/3 every 2 to 3 days until either a satisfactory maintenance level has been achieved or therapy can be discontinued. In acute or self-limiting disorders, treatment may be discontinued as soon as control has been obtained.

SUPPLIED: GAMMACORTEN Tablets, 0.75 mg. 2/2704 MR

BEFORE GAMMACORTEN: **M. S. could not raise arms above shoulder level; even the degree of motion shown was extremely painful.**

AFTER ONE WEEK OF GAMMACORTEN: **Range of motion and rotation dramatically increased; M. S. could move arms without pain for the first time in months.**

Gammacorten T.M.

(dexamethasone CIBA)

...a potent, highly effective corticosteroid; profound anti-inflammatory activity, with minimal potential for corticosteroid side effects

CIBA
SUMMIT, N. J.

new for total management of itching,[1,2] inflamed,[3,4] infected[5,6] skin lesions

Dermatitis repens with staph and monilia 7 weeks duration

Cleared in 5 days

Mycolog ointment

Kenalog, Spectrocin and Mycostatin in Plastibase

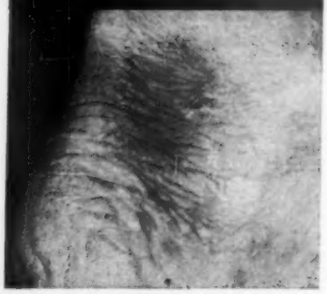
antipruritic/anti-inflammatory/antibacterial/antifungal

Mycolog Ointment – containing the new superior topical corticoid Kenalog – reduces inflammation,[3,4] relieves itching,[1,2] and combats or prevents bacterial, monilial and mixed infections.[5-7] It is extremely well tolerated, and assures a rapid, decisive clinical response for most infected dermatoses.

> **"Thirty-one of 38 patients . . . obtained excellent or good control of dermatological lesions . . . [Mycolog] was highly effective, particularly in the management of mixed infections. Several recalcitrant eruptions which had not responded to previous therapy were remarkably responsive to the daily application of this preparation over periods of 2 to 3 weeks."[7,8]**

For total management of itching, inflamed, infected skin lesions, Mycolog contains triamcinolone acetonide, an outstanding new topical corticoid for prompt, effective relief of itching, burning and inflammation[1-4] – neomycin and gramicidin for powerful antibacterial action[7] – and nystatin for treating or preventing Candida (Monilia) albicans infections.[5,9]

Infectious eczematoid dermatitis of ankle–5 years duration



Cleared in 20 days

Application: Apply 2 to 3 times daily. Supply: 5 Gm. and 15 Gm. tubes. Each gram supplies 1.0 mg. (0.1%) triamcinolone acetonide, 2.5 mg. neomycin base, 0.25 mg. gramicidin, and 100,000 units nystatin in PLASTIBASE.

References: 1. Shelmire, J.B., Jr.: Monographs on Therapy 3:164 (Nov.) 1958. · 2. Nix, T.E., Jr., and Derbes, V.J.: Monographs on Therapy 3:123 (Nov.) 1958. · 3. Robinson, R.C.V.: Bull. School of Med., U. Maryland 43:54 (July) 1958. · 4. Sternberg, T.H.: Newcomer, V.D., and Reisner, R.M.: Monographs on Therapy 3:115 (Nov.) 1958. · 5. Clark, R.F., and Hallett, J.J.: Monographs on Therapy, 3:153 (Nov.) 1958. · 6. Smith J.G., Jr.; Zawisza, R.J., and Blank, H.: Monographs on Therapy, 3:111 (Nov.) 1958. · 7. Monographs on Therapy, 3:137 (Nov.) 1958. · 8. Howell, C.M., Jr.: North Carolina M.J. 19:449 (Oct.) 1958. · 9. Bereston, E.S.: South. M.J. 50:547 (April) 1957.

And whatever the topical corticoid need, a suitable Squibb formulation is available—Kenalog-S Lotion—7½ cc. plastic squeeze bottles. Each cc. supplies 1.0 mg. (0.1%) triamcinolone acetonide, 2.5 mg. neomycin base and 0.25 mg. gramicidin. Kenalog Cream, 0.1%—5 Gm. and 15 Gm. tubes. Kenalog Lotion, 0.1%—15 cc. plastic squeeze bottles. Kenalog Ointment, 0.1%—5 Gm. and 15 Gm. tubes.

RELIABILITY · SQUIBB

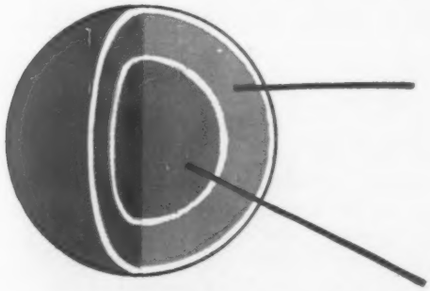
SQUIBB

Squibb Quality — the Priceless Ingredient

'SPECTROCIN'®, 'MYCOSTATIN'®, 'PLASTIBASE'®, 'MYCOLOG' AND 'KENALOG' ARE SQUIBB TRADEMARKS

the complaint: "nervous indigestion"

the diagnosis: any one of several nonspecific gastrointestinal disorders requiring relief of symptoms by sedative-antispasmodic action with concomitant digestive enzyme therapy. **the prescription:** a new formulation, incorporating in a single tablet the actions of Donnatal and Entozyme. **the dosage:** two tablets three times a day, or as indicated.



the formula: *in the gastric-soluble outer layer:*

Hyoscyamine sulfate	0.0518 mg.
Atropine sulfate	0.0097 mg.
Hyoscine hydrobromide	0.0033 mg.
Phenobarbital (1/8 gr.)	8.1 mg.
Pepsin, N.F.	150 mg.

in the enteric-coated core:

Pancreatin, N.F.	300 mg.
Bile salts	150 mg.

DONNAZYME™ Rx Robins

A. H. ROBINS COMPANY, INCORPORATED • RICHMOND 20, VIRGINIA

V-CILLIN K®. . .

dependable, fast, effective therapy

V-Cillin K produces therapeutic blood levels in all patients within five to fifteen minutes after administration—levels higher than those attained with any other oral penicillin. Infections resolve rapidly. *Dosage:* 125 or 250 mg. three times daily. *Supplied:* In scored tablets of 125 and 250 mg. (200,000 and 400,000 units).

New: V-Cillin K® Sulfa. Each tablet combines 125 mg. of V-Cillin K with 0.5 Gm. of the three preferred sulfonamides.

New: V-Cillin K, Pediatric, a taste treat for young patients. In bottles of 40 and 80 cc. Each 5-cc. teaspoonful provides 125 mg. of V-Cillin K.

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Editorial

The Systemic Mycoses

REDUCTION and control of microbial diseases through the discovery and use of specific therapeutic antimicrobial substances has now been largely accomplished. With the exception of resistant staphylococcal infections, it has been possible to check spread of the decimating bacterial diseases, such as plague, cholera, typhoid, salmonellosis and many others. In the treatment of filterable virus diseases, the number of effective therapeutic drugs available is much smaller, but prevention with vaccines has proved entirely feasible for some of these diseases. Patients afflicted with disseminated mycotic disease, on the other hand, have been compelled to struggle through the natural course of the disease without the aid of any really effective chemotherapeutic agents. This situation began to improve following the introduction of sulfonamides, penicillin and broad-spectrum antibiotics for the treatment of actinomycosis and nocardiosis; the use of hydroxystilbamidine for North American blastomycosis; and especially since the use of amphotericin B for almost all of the systemic mycoses.

Systemic mycotic infections in the United States fall into one of the following categories: (1) Primary infections, such as coccidioidomycosis, histoplasmosis, North American blastomycosis, cryptococcosis, moniliasis, sporotrichosis, actinomycosis and nocardiosis. The incidence of the first three infections varies with the geographical regions of the country. (2) Secondary infections, usually clinical variations of moniliasis. These emerge as complications during the treatment of bacterial or viral infections with broad-spectrum antibiotics. Cases of this type are now fairly numerous and have a uniform geographic distribution. (3) Secondary infections, such as cryptococcosis, histoplasmosis, mucormycosis, aspergillosis and moniliasis. These appear as complications in persons suffering from primary debilitating diseases, such as Hodgkin's disease, lymphosarcoma, leukemia, sarcoidosis, tuberculosis or other primary mycoses, and emerge particularly among those who may have received treatment with corticosteroids, nitrogen mustard or antibiotics. In many instances, fatalities ensue from invasion of the tissues by the pathogenic fungi rather than as a result of the primary disease [*1*].

Despite improvements in diagnostic procedures, recognition of systemic mycotic disease still is governed more by the availability of laboratory personnel experienced in the required technical procedures than by any other factor.

Isolation of the etiologic agent of systemic mycoses from the soil and determination of endemicity by skin testing have revealed ever widening regions of infection. Studies now show that the area of endemicity of histoplasmosis, formerly believed to be confined to the central Mississippi Valley, now extends over the entire eastern United States from the Great Lakes Region, and encompassing New York, Washington, D. C. and Louisiana. Recently Furcolow described the spreading of the mold parasite Histoplasma capsulatum by tornadoes [*2*]. In Cincinnati, Schwarz and associates [*3*] detected splenic calcifications, attributable to histoplasmosis, in almost half of routine autopsy cases. Coccidioidomycosis, while sharply localized to California, is found throughout the

Southwest and has appeared in individuals who have made even brief visits to these regions. The etiologic agent of cryptococcosis, Cryptococcus neoformans, first isolated from the soil by Emmons in 1951 [4], subsequently was recovered by him from pigeon droppings in Virginia [5]. The parasite again was encountered in pigeon excretions in Cincinnati by Kao and Schwarz [6], and was found in widely scattered states by Ajello [7]. It was detected in high incidence in pet shops and pigeon coops in New York City by Littman and Schneierson [8]. Pulmonary nocardiosis, caused by Nocardia asteroides, a highly malignant soil organism, is being encountered with increasing frequency. There is also an increase in occurrence of aspergillosis appearing as a primary pulmonary disease and as a complication particularly in patients with diabetes mellitus. Epidemics of North American blastomycosis and histoplasmosis have recently been reported.

Because of the cross-reactions between fungal antigens, it is common practice to skin test individuals with a battery of antigens; e.g., coccidioidin, histoplasmin, blastomycin and tuberculin, and to examine the site daily for skin hypersensitivity. At the present time there is no satisfactory skin test for cryptococcosis. Despite the usefulness of serologic technics, such as complement fixation, precipitin and agglutinations tests, an exact diagnosis can be made only by isolation and identification of the etiologic agent. Nevertheless, serologic evidence of the disease usually is the first to bring attention to the possible diagnosis of mycotic infection.

Improvements in laboratory diagnostic methods have progressed to the point where the majority of pathogenic fungi can be isolated directly from a patient's exudates within three to six days. Culture of gastric aspirate of patients with pulmonary mycotic infections is especially valuable for recovering the etiologic agent. Instead of using the slow animal technic of inoculation of white Swiss mice with exudates, followed by cultivation of the animal's liver and spleen on peptone agars, a process that may take from three to six weeks, liver-spleen-glucose-blood agar [9] has been successfully employed for the rapid direct isolation of pathogenic fungi at 37°c. Identification of these fungi is still dependent upon their morphologic characteristics; however, studies of the nutritional and biochemical characteristics of pathogenic fungi are proceeding rapidly. Thus the discovery that C. neoformans possesses an absolute requirement for thiamine and its moieties, and is capable of assimilating only members of the glutamic acid family, furnished a rational for withholding thiamine and vitamin B complex in the treatment of patients with cryptococcal meningitis [10]. The disseminated nature of cryptococcosis in reticuloendothelial diseases such as leukemia, Hodgkin's disease and lymphosarcoma is thought to be due to specific changes in the nutritional content of blood cells and plasma that favor the growth of C. neoformans. Studies of the nutritional requirements of Coccidioides immitis, H. capsulatum and Candida albicans also have revealed useful information.

With the discovery of amphotericin B (Fungizone®) [11] and its use as an intravenous medication, treatment of the systemic mycoses has been greatly improved. The consensus of investigators is that amphotericin B is highly effective for disseminated mycoses such as North American blastomycosis, South American blastomycosis, histoplasmosis, coccidioidomycosis, cryptococcosis and moniliasis. Azotemia occurs when the dose is excessive or too frequent, and administration therefore is limited to alternate days. No permanent adverse effects on the heart, liver, bone marrow, central nervous system or skin have been observed. Side effects include nausea, occasional vomiting, flushing, perspiration, fatigue, drowsiness, febrile reactions, anxiety and generalized pain, which usually subside upon discontinuation of the medication. When given intravenously, the antibiotic can produce a blood level of 2 μg./ml., which fortunately exceeds the minimal inhibitory concentration of most pathogenic fungi. For example, a patient with leukemia complicated by monilial or cryptococcal dissemination can be cleared of the fungal parasites with a relatively small number of infusions. In the treatment of disseminated mycotic diseases such as coccidioidomycosis, histoplasmosis and North American blastomycosis, however, therapy must be prolonged for several months, during which time a minimum of 2,000 to 3,000 mg. is administered. The treatment of coccidioidal and cryptococcal meningitis also must be prolonged, since amphotericin B fails to cross the blood-brain barrier in sufficient concentration to effect rapid eradication of the fungus. Besides the intravenous route, the antibiotic has been administered intrathecally, intra-articularly, intrathoracically, and by aerosol inhalation. It has also been

applied successfully to the conjunctiva, and injected into the anterior chamber of the eye in the treatment of fungal endophthalmitis [*12*]. It has been used successfully by subcutaneous injection for the local treatment of chromoblastomycosis [*13*] and by injection for the treatment of fungal empyemas [*14*]. When given orally amphotericin B is absorbed only to a slight degree and does not yield appreciable blood levels of the drug.

The clinical effectiveness of amphotericin B for the systemic mycoses is limited by the rapidity with which susceptible pathogenic fungi become resistant to the antibiotic. Evidence is accumulating that candida species may become resistant to polyene antibiotics, including nystatin and amphotericin B. Stout and Pagano [*15*] observed that one of their five strains of C. albicans exhibited a threefold increase in resistance to nystatin. Littman, Pisano and Lancaster [*16*] failed to induce resistance in one strain of C. albicans, but accomplished it with other members of candida genus. Sorensen and associates [*17*] produced a strain of C. albicans resistant to high concentrations of amphotericin B, and cross resistant to nystatin. It is clear that C. albicans can become resistant to amphotericin B and that the strain may also be cross resistant to mystatin. Although the need for prolonged therapy of the systemic mycoses seems to favor the development of resistance to amphotericin B, preliminary evidence that C. immitis does not become readily resistant to amphotericin B is found in the fact that isolates of the organism, recovered after thirty-two intravenous doses of antibiotic had been administered, were as sensitive to amphotericin B as the first isolate [*1*]. Despite this, Sorensen and associates [*17*] were able to develop two strains of C. immitis that exhibited a tenfold degree of resistance to amphotericin B. The current wide use of nystatin together with broad-spectrum antibiotics as a prophylactic to prevent monilial overgrowth in the gastrointestinal tract during antibiotic therapy may very well lead to the increased resistance of C. albicans and other pathogenic fungi to both nystatin and amphotericin B. This could be disastrous for patients with disseminated mycoses, since amphotericin B is the only really effective medication available at the present time. In the treatment of actinomycosis and nocardiosis, amphotericin B is ineffective. However, both these diseases respond to protracted treatment with penicillin, sulfadiazine and broad-spectrum antibiotics.

The treatment of systemic mycoses frequently involves other forms of therapy. For example, coccidioidal cavities in the lung do not close after administration of amphotericin B and surgery has a definite place in the therapy of this disease. An absolute indication for pulmonary resection of patients with coccidioidomycosis is the presence of giant cavities of more than 5 cm. in size, infected or ruptured cavities, the presence of bronchopleural fistulas, empyema, non-expansile lung, and persistent pneumothorax [*18*]. Since the osseous system is frequently involved, and draining sinuses are common, surgical débridement and immobilization of affected extremities are necessary adjuncts to treatment.

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Clinical Studies

Clinical Characteristics of Leptospirosis*

Observations Based on a Study of Twelve Sporadic Cases

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McKinney, Texas

EARLY discussions [*1–3*] of leptospirosis dealt primarily with Weil's disease, a severe clinical syndrome characterized by fever, jaundice, renal damage and hemorrhage. In recent years, however, attention has been drawn, particularly by observers in Europe [*4*] and Asia [*5*], to the fact that leptospirosis is usually a mild disease in which jaundice and significant renal damage are unusual, and that this mild form can result from infection with any of the leptospiral serotypes.

In the following study twelve cases of leptospirosis are reported. Examination of the clinical material discloses three salient features: (1) different serotypes produce the same mild disease; (2) the clinical course can be divided into two distinct phases; and (3) the manifestations in the first stage of leptospirosis are due to leptospiremia, while those in the second stage appear to be the consequence of the body's immunologic response.

CASE REPORTS

CASE 1. L. L., a thirty-three year old Negro bartender from Denison, Texas, entered the McKinney Veterans Administration Hospital on June 14, 1957. He had had abrupt onset of fever, recurrent chills, frontal headache, myalgia, abdominal pain, malaise, anorexia and constipation seventy-two hours prior to admission. Symptoms had persisted despite tetracycline therapy, which had been started on the second day, and his physician had advised hospitalization. He denied excessive intake of alcohol. In the two weeks prior to the onset of illness he had been exposed to a sick dog and had waded through stagnant water while fishing.

The temperature was 103.6°F.; pulse, 112; and blood pressure, 115/80 mm. Hg. Conjunctival suffusion, stiffness of the neck, muscular tenderness, abdominal tenderness, distention and hypoactive peristaltic sounds were the only abnormal physical findings.

The hemoglobin was 13.5 gm./100 cc.; erythrocyte sedimentation rate, 32 mm. (Wintrobe); white blood cell count, 7,600/cu. mm.; and neutrophils, 76 per cent. The urine specific gravity was 1.025, there was 2-plus protein, and a few granular casts and white cells per low power field on microscopic examination. Liver function tests were normal on the sixth day, but on the ninth day the serum bilirubin was 1.5 mg./100 cc. and thymol turbidity 10 units.

The course of illness is shown in Figure 1. Because of headache and stiff neck a lumbar puncture was performed shortly after admission but the cerebrospinal fluid was normal. The patient's condition changed little until the morning of the sixth day when his behavior became quite abnormal. He ran down the hall screaming, refused to take medications, disappeared from his room and was found at a restaurant 400 yards away, and beat a ward attendant severely about the face. Three days later his delirium cleared and in a short time he was asymptomatic. A repeat lumbar puncture on the sixth day revealed 11 cells/cu. mm. Conjunctival suffusion persisted for a week following admission.

The agglutination-lysis test revealed agglutinins for several serotypes on June 28, but on July 16 the only significant titer was for Leptospira canicola. An attempt to culture leptospirosis from blood drawn on the fourth day was unsuccessful.

Comment: This patient was moderately ill with severe delirium and mild disturbance of his liver function tests. There was good presumptive evidence that his infection was due to L. canicola, an organism which some authors consider noteworthy because of the high incidence of delirium it causes [*6*]. McCrumb found many patients with similar slight disturbances of liver function tests [*5*].

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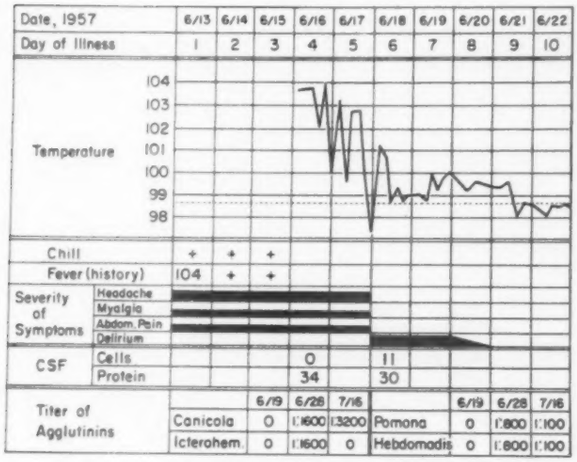


FIG. 1.

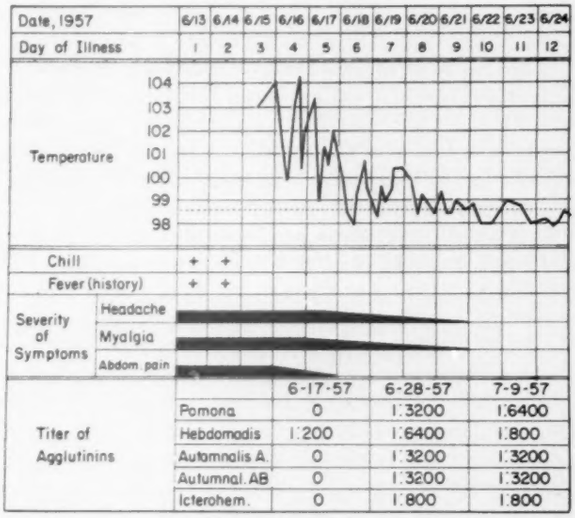


FIG. 2.

In this report and those to follow, the results of routine blood and urine cultures, chest roentgenograms, serological tests for syphilis, agglutination tests for common pathogenic bacteria, and spinal fluid smears, cultures, Wassermann reactions and colloidal gold reactions were negative unless otherwise indicated.

CASE II. R. V., a sixty-two year old white farmer from Van Alstyne, Texas, entered the McKinney Veterans Administration Hospital on June 16, 1957. He became ill on the morning of June 13 with fever and stiffness, soreness and aching of his legs, and that evening experienced the first of several chills. Thereafter he had high fever, severe frontal headache, constipation, anorexia, malaise and lower abdominal pain, but no cough, rash, jaundice or diarrhea. He kept pigs on his farm and his only cow had died of an undiagnosed disease approximately two weeks before the onset of his illness. He did not recall contact with rodents or stagnant water.

On admission he appeared acutely ill. The temperature was 103°F.; pulse, 90; and blood pressure, 160/86 mm. Hg. There was suffusion of the conjunctivas and tenderness and distention of the abdomen, but no jaundice or nuchal rigidity. The hemoglobin was 17.8 gm./100 cc.; erythrocyte sedimentation rate, 14 mm. (Wintrobe); white blood cell count, 8,900/cu. mm.; and neutrophils, 79 per cent. The urine was normal on admission but three days later contained 2-plus protein. Two days following admission the erythrocyte sedimentation rate was 32 mm. There was 3-plus cephalin-cholesterol flocculation but results of all other liver function tests were normal. The blood urea nitrogen was 26 mg./100 cc. on admission but on subsequent determinations it was normal.

The course of illness is shown in Figure 2. There was defervescence and improvement in symptoms on the fifth day, but the patient continued to complain of muscle aching and a feeling of discomfort in the bones until the fourteenth day. No symptoms or signs of meningitis were detected and the spinal fluid was not examined. A faint, blotchy, erythematous rash appeared on the sixth day but vanished overnight.

Agglutination-lysis tests on serial serum specimens revealed rising titers of agglutinins for several leptospiral serotypes. Blood cultured on the fourth day, using Korthof's medium, was positive for L. pomona.

Comment: This man's course was unusual in that myalgia and bone discomfort persisted for so long. Although he had L. pomona infection, proved by isolation of the organism, the agglutination-lysis test failed to give unequivocal evidence of infection by that organism. Even though he had no signs or symptoms of meningitis, it should not be concluded that his spinal fluid was normal.

CASE III. C. B., a thirty year old white construction worker from Blue Ridge, Texas, entered the McKinney Veterans Administration Hospital on July 17, 1957, with an illness which began rather insidiously four days prior to admission with generalized muscular aches, headache, fever and slight nausea. He was treated with penicillin from the first day of illness but did not improve, and was referred for hospitalization for an "influenzal illness with fever and muscle aches." He did not recall contact with animals of any kind but had worked around stagnant water on a highway construction project near McKinney, Texas.

He arrived on the hospital ward while having a chill. The temperature was 104.4°F.; pulse, 100; and blood pressure, 130/80 mm. Hg. There was conjunctival suffusion and mild nuchal rigidity, but no icterus or rash. The hemoglobin was 14.6 gm./100 cc.;

white blood cell count, 6,800/cu. mm.; and neutrophils, 85 per cent. The urine contained 1-plus protein.

The course of illness is shown in Figure 3. Meningitis was suspected on admission but the spinal fluid was normal. On the sixth day he was much improved, even though headache and nuchal rigidity were still present and he had begun to complain of photophobia. In the early morning of the seventh day he was awakened by an intense headache, and on examination was noted to have increased conjunctival suffusion and nuchal rigidity. At 4:30 P.M. a spinal puncture revealed elevated pressure (220 mm. of fluid), 288 cells/cu. mm. (75 per cent polymorphonuclears) and protein 140 mg./100 cc. The headache improved following the spinal puncture, and the next day only slight headache, stiffness of the neck, conjunctivitis and photophobia remained. He was asymptomatic from the ninth day, even though pleocytosis was still present on the sixtieth day. Serial agglutination-lysis tests revealed rising titers of agglutinins for L. pomona, and lower titers for L. autumnalis A and L. autumnalis AB.

Comment: This relatively mild case illustrates the occurrence in the first stage of meningismus in the presence of normal spinal fluid, the abrupt onset of clinical meningitis on the seventh day and prolonged persistence of pleocytosis. The agglutination-lysis test permitted presumptive identification of the infecting serotype as L. pomona.

CASE IV. J. C., a twenty-three year old white farmer from Allen, Texas, was admitted to the McKinney Veterans Administration Hospital on July 29, 1957. He became ill on July 25 with severe frontal headache, cramping lower abdominal pain, nausea, chills and fever. Tetracycline therapy was begun on the second day but he remained acutely ill with recurrent chills, fever, headache, myalgia, vomiting, prostration, anorexia and constipation. He kept

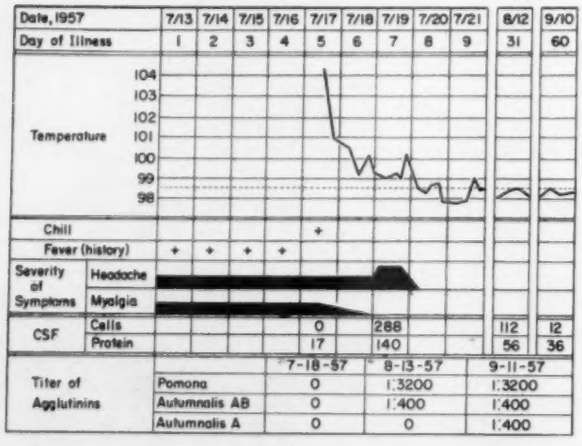


FIG. 3.

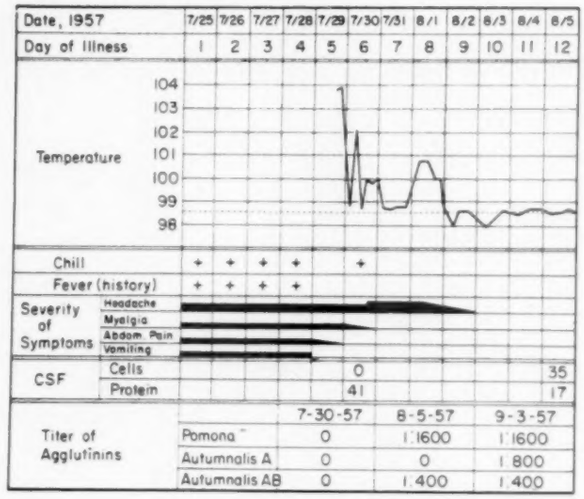


FIG. 4.

a cat and pigs on his farm, and on July 18 he swam in a pond frequented by a variety of animals.

He was acutely ill with temperature of 103.6°F.; pulse, 96; and blood pressure, 116/76 mm. Hg. There was conjunctival injection and a palpable spleen, but no icterus, stiffness of the neck or rash. The hemoglobin was 15 gm./100 cc.; erythrocyte sedimentation rate, 35 mm. (Wintrobe); white blood cell count, 4,400/cu. mm.; and neutrophils, 73 per cent. The urine was normal.

The course of illness is shown in Figure 4. The first stage terminated on the sixth day with fall in temperature and subsidence of all symptoms except headache. The spinal fluid was normal on the morning of the sixth day. In the late afternoon of the sixth day there was abrupt exacerbation of headache. This persisted for three days before he became asymptomatic. A repeat lumbar puncture on the twelfth day revealed 35 cells/cu. mm. (31 lymphocytes and 4 neutrophils) and normal protein and sugar. Serological tests revealed rising titers of agglutinins for several serotypes of leptospires.

Comment: This case illustrates the abrupt onset of clinical meningitis on the sixth day, after a fairly typical first stage. It is interesting that symptoms of meningitis developed less than twelve hours after a negative lumbar puncture, but unfortunately the spinal fluid was not examined again until six days later. His temperature curve was of the classic biphasic type. The diagnosis of leptospirosis was confirmed serologically but identification of the organism as L. pomona was questionable.

CASE V. J. P., a forty-eight year old white hospital ward attendant and farmer from McKinney, Texas, became acutely ill on August 13, 1957, with fever and

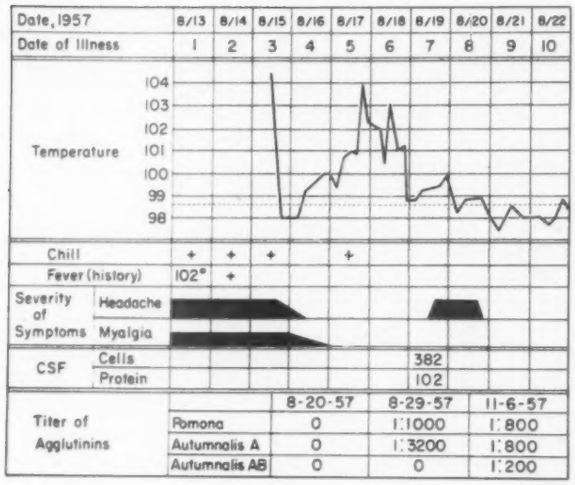


FIG. 5.

aching legs. Later he had a chill, and suboccipital throbbing headache and soreness of the neck muscles developed. These symptoms persisted until his admission to the McKinney Veterans Administration Hospital on August 15. One of the cows on his farm had been sick in the spring and a veterinarian had diagnosed leptospirosis. Approximately a week before onset of illness he swam in a pond used by his livestock for water.

The temperature was 104.4°F.; pulse, 116; and blood pressure, 120/80 mm. Hg. There was conjunctival injection and a palpable liver edge, but no icterus, rash or nuchal rigidity. The hemoglobin was 14.7 gm./100 cc.; erythrocyte sedimentation rate, 33 mm. (Wintrobe); white blood cell count, 13,000/cu. mm., and neutrophils, 92 per cent. The urine was normal.

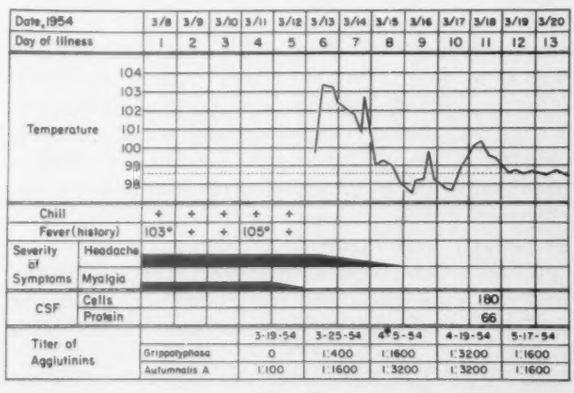
The course of illness is shown in Figure 5. His temperature fell on the day of admission, following the administration of 0.6 gm. of aspirin, and headache and myalgia subsided. However, on the fifth day he had a chill and a spike in temperature, but there were no specific complaints other than anorexia and weakness. The administration of tetracycline, 2 gm. daily, was started at that time. On the seventh day he had abrupt recurrence of severe throbbing headache which was not relieved by salicylates. Although his neck was supple, a lumbar puncture performed soon after onset of headache revealed cerebrospinal fluid under normal pressure but containing 382 cells/cu. mm. (91 per cent polymorphonuclears). Headache subsided the next day and he was asymptomatic after the ninth day. There were rising titers of agglutinins for both L. pomona and L. autumnalis A. On December 19, 1957, the agglutinin titers were 1:400 for both serotypes.

Comment: The course in the first stage was somewhat unusual in that he had little fever on the fourth day, and headache and myalgia subsided earlier than in other patients and did not recur with the second febrile period on the fifth and sixth days. The fever again subsided on the sixth day and clinical meningitis appeared on schedule. The significance of the fall in temperature on the third day is not clear although it might have marked the end of the first stage. Gsell [*4*] described cases in which the initial febrile period terminated on the third or fourth day. Identification of the infecting serotype was impossible because of parallel titers for two organisms.

CASE VI. O. D., a sixty-two year old white farmer from Wolfe City, Texas, entered the McKinney Veterans Administration Hospital on March 3, 1954, five days after abrupt onset of chills, fever (105°F.), headache (bitemporal, dull, aching and sometimes throbbing), myalgia, arthralgia and malaise. He had been given "fever pills" by his physician, but no antibiotics. He had been exposed to cats and horses on his farm and had killed squirrels in the month preceding his illness. There was no history of contact with rats, mice or stagnant water.

He was acutely ill with temperature of 99.6°F.; pulse, 112; and blood pressure, 130/70 mm. Hg. On admission no conjunctival injection, nuchal rigidity, icterus, rash or other abnormal physical findings were noted. The hemoglobin was 13.5 gm./100 cc.; erythrocyte sedimentation rate, 35 mm. (Wintrobe); white blood cell count, 8,500; and neutrophils, 97 per cent. The urine was normal; blood urea nitrogen, 42 mg./100 cc.; serum CO_2 combining power, 24 mEq./L.; and serum chloride, 98 mEq./L.

The course of illness is shown in Figure 6. There was defervescence and asymptomatic improvement on the seventh day. The diagnosis of leptospirosis was first suspected on the ninth day when conjunctival suffusion was noted and a history of photophobia was elicited. Although he had no clinical evidence of meningitis, a lumbar puncture performed on the



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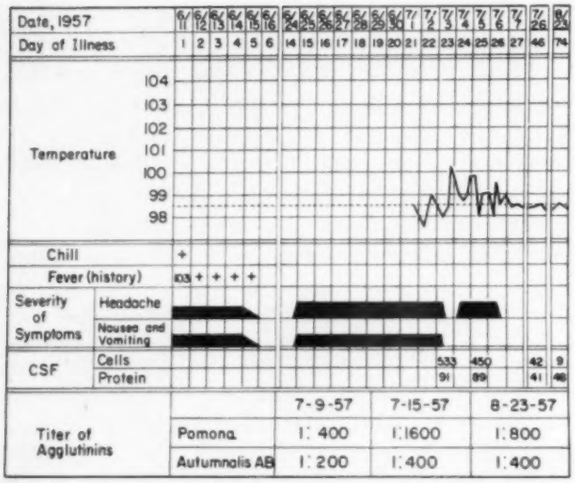

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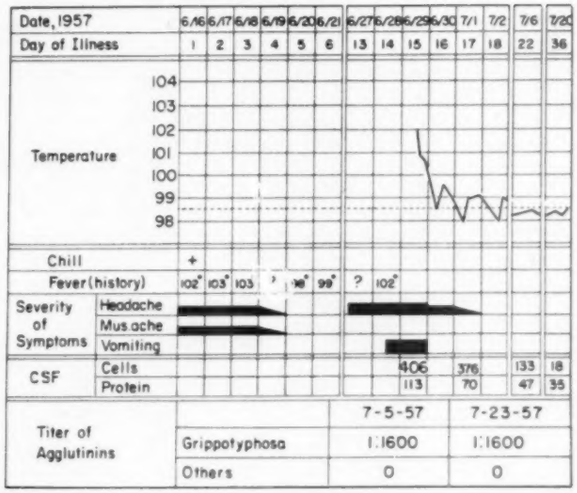

FIG. 8.

eleventh day revealed 180 cells/cu. mm. (66 per cent polymorphonuclears) and a pressure of 200 mm. of fluid. The blood urea nitrogen soon fell to normal levels, and on March 11 there was 20 per cent phenolsulfonephthalein excretion in fifteen minutes. He was asymptomatic after the eleventh day. Agglutination-lysis tests revealed rising titers of agglutinins for both L. grippotyphosa and L. autumnalis A.

Comment: This case illustrates the importance of conjunctival suffusion and pleocytosis as diagnostic clues, as these findings led to the correct diagnosis. He had no symptoms of meningitis in the second stage, yet there was a definite meningeal reaction. The mild elevation of blood urea nitrogen was probably extrarenal in origin since there was no abnormality on urinalysis. Identification of the infecting serotype from the results of the agglutination-lysis tests was not possible.

CASE VII M. V., a thirty year old white male school teacher from Allen, Texas, entered the McKinney Veterans Administration Hospital on July 1, 1957, with headache, nausea and vomiting of six days' duration. Three weeks prior to admission he had been ill for four or five days with a chill, fever, headache, nausea and vomiting. This illness was diagnosed as "the flu" and he was treated with tetracycline. He then felt well enough to return to work, but on June 24 (fourteenth day of illness) severe frontal headache, blurring of vision, nausea and vomiting suddenly developed; there was no fever, chills or myalgia. He admitted swimming in a small stream, drinking water from an abandoned well and caring for a sick dog during the two weeks prior to onset of his initial symptoms.

His blood pressure was 130/90 mm. Hg; pulse, 70; and temperature, 98.6°F. The only abnormal physical findings were stiffness of the neck and conjunctival suffusion. The hemoglobin was 16.9 gm./100 cc.; erythrocyte sedimentation rate, 9 mm. (Wintrobe); white blood cell count, 8,000 cu. mm.; and neutrophils, 66 per cent. The urine was normal.

The course of illness is shown in Figure 7. Cessation of vomiting and temporary relief of headache followed a lumbar puncture on July 3. The spinal fluid pressure was 410 mm. of fluid; cell count, 533/cu. mm. (98 per cent lymphocytes); protein, 91 mg./100 cc.; and sugar, 65 mg./100 cc. Because of low grade fever he was given tetracycline, 2 gm. per day for four days, beginning July 3. The headache returned twenty-four hours after the first lumbar puncture but subsided after another forty-eight hours and he was asymptomatic thereafter, even though his cerebrospinal fluid still contained 9 cells/cu. mm. seventy-four days after onset of illness. There was a fourfold rise in agglutinin titers for L. pomona and a twofold rise for L. autumnalis AB.

Comment: In contrast to the preceding patients, this patient and the next patient were admitted in the second stage of illness with the symptoms and findings of "aseptic meningitis" rather than septicemia. The initial illness in each case was relatively mild and could easily have been overlooked or dismissed as being unrelated to the illness causing hospitalization. Both cases were unusual in that the onset of symptoms of meningitis was delayed until the end of the second week. Gsell [*7,8*] has described two such patients, one with meningitis appearing on the fourteenth day and persisting beyond the forty-ninth day, and a second with cells appearing on the twelfth day and persisting beyond the eighty-fourth day. Gsell [*9*] believes that the late onset

of meningitis is related to inadequate antibiotic therapy in the first stage. Case VII can be explained on this basis, but in Case VIII there was no antibiotic therapy prior to admission. The febrile episode beginning on the twenty-second day presumably represented a mild relapse since no other cause for fever was found. This patient probably had L. pomona infection but the agglutination-lysis results were somewhat equivocal.

CASE VIII. R. M., a forty-one year old white truck driver, entered the McKinney Veterans Administration Hospital on June 29, 1957. His illness began two weeks prior to admission with fever, headache, myalgia and a chill. Although his symptoms had already subsided, he saw a physician on the fifth day and was hospitalized in a private hospital for the next four days. During that time his maximum temperature was 99°F. and he complained only of slight malaise. He felt relatively well until the thirteenth day of illness when he awakened with intense bitemporal headache, retrobulbar pain and a stiff, painful neck. On the following day he had onset of fever and frequent vomiting. He denied contact with animals of any kind but had waded in a small stream near Gainesville, Texas, a few days prior to onset of illness.

The temperature was 102°F.; pulse, 92; and blood pressure, 140/90 mm. Hg. He was acutely uncomfortable but well oriented. There was moderate nuchal rigidity, without Kernig's or Brudzinski's signs and the eyes were normal. The hemoglobin was 14.4 gm./100 cc.; erythrocyte sedimentation rate, 28 mm. (Wintrobe); white blood cell count, 13,900/cu. mm.; and neutrophils, 85 per cent. The urine was normal.

The course of illness is shown in Figure 8. Lumbar puncture on admission disclosed a pressure of 350 mm. of fluid; 406 cells/cu. mm. (95 per cent lymphocytes); protein, 113 mg./100 cc.; and sugar, 69 mg./100 cc. Vomiting ceased after the first lumbar puncture but the headache persisted for two days before he became asymptomatic. Tetracycline therapy, 2 gm. per day, was begun on the day of admission and continued for eight days. His serum agglutinated L. grippotyphosa in a sustained titer of 1:1600.

Comment: There was little to suggest leptospirosis in this patient except the short febrile illness beginning two weeks prior to admission. The clinical pictures in Cases VII and VIII were remarkably similar even though the infections were due to different serotypes. The febrile episode beginning on the fourteenth day was thought to be a relapse.

CASE IX.* C. R., a thirty-seven year old farmer from Celina, Texas, entered the McKinney Veterans Administration Hospital on September 9, 1950, on the fourth day of illness. The symptoms were abrupt in onset, with recurrent chills, temperature elevation to 104°F., headache, aching of all joints, nausea, vomiting, retrobulbar pain, abdominal pain and a few loose bowel movements. He had noted redness of his eyes for two or three days prior to admission. Various unidentified oral medications had been taken without benefit. He kept cows and pigs on his farm, had noted rats in his barn, and on several occasions had assisted a veterinarian in caring for sick animals.

He appeared acutely ill with temperature 103°F.; pulse, 108; and blood pressure, 105/70 mm. Hg. The conjunctivas were suffused and there was upper abdominal tenderness, but no icterus, rash or nuchal rigidity. The hemoglobin was 17.5 gm./100 cc.; erythrocyte sedimentation rate, 39 mm. (Wintrobe); white blood cell count, 8,750/cu. mm.; and neutrophils, 84 per cent. The urine contained 1-plus protein.

On the fifth day of illness there was defervescence and disappearance of all symptoms except weakness. He remained afebrile on the sixth day but on the seventh day his temperature rose to 102°F. and remained elevated for the next forty-eight hours. After the recurrence of fever he was given 2 gm. of Chloromycetin® daily for six days. Lumbar puncture on the eighth day revealed 13 cells/cu. mm.; protein, 45 mg./100 cc.; sugar, 89 mg./100 cc.; and pressure of 180 mm. of fluid. He was free of symptoms after the ninth day.

Agglutinin titers for L. pomona were 1:3200 on the ninth day, 1:6400 on the sixteenth day, and 1:100 on the eightieth day. There was no agglutination of L. canicola and L. icterohemorrhagiae, the only other antigens used.

Comment: This patient had a relatively mild illness with asymptomatic meningitis and typical biphasic temperature curve. He was the only patient with arthralgia as a major symptom. He also exhibited mild diarrhea, an infrequent complaint. The high agglutinin titers for L. pomona were considered strong presumptive evidence of infection by that serotype.

CASE X.* B. G., a fifty-seven year old farmer from Howe, Texas, entered the McKinney Veterans Administration Hospital on October 2, 1950, on the seventh day of illness. The symptoms were abrupt in onset, with a chill, fever (103°F.), headache, aching of the lumbar muscles and malaise. His physician gave him penicillin and sulfonamide on the second day, and thereafter he felt better until the sixth day when he had another chill and exacerbation of headache, fever and malaise, and the onset of a nonproductive cough. He kept cats, dogs, cows and swine

* Previously reported [*10*].

* Previously reported [*10*].

on his farm and frequently worked as a veterinarian's assistant.

He was acutely ill and dehydrated on admission. The temperature was 102°F.; pulse, 84; and blood pressure, 110/60 mm. Hg. Scattered coarse rales were heard over both lung bases posteriorly. There was no jaundice, conjunctival injection, rash or nuchal rigidity.

The hemoglobin was 13 gm./100 cc.; erythrocyte sedimentation rate, 33 mm. (Wintrobe); white blood cell count, 12,600/cu. mm.; and neutrophils, 80 per cent. The urine contained 1-plus protein and a few granular casts and white cells per low power field. Roentgenographic examination of the chest demonstrated a small irregular infiltration in the lower right lung field. Results of liver function tests, performed on the twenty-sixth day, were normal except that the thymol turbidity was 15 units and serum globulin 3.8 gm./100 cc. (albumin, 4.0 gm./100 cc.). On February 19, 1951, the total protein was 6.5 gm./100 cc.; globulin, 2.1 gm./100 cc.; and thymol turbidity, 4 units.

The temperature rose to 104°F. soon after admission, fell below 101°F. on the eighth day, and then fluctuated between normal and 100.6°F. for the next three days. Backache and other complaints subsided after the seventh day, but headache persisted until the fifteenth day. Lumbar puncture on the tenth day revealed 56 cells/cu. mm. (76 per cent polymorphonuclears), protein, 82 mg./100 cc.; sugar, 63 mg./100 cc.; colloidal gold reaction, 4433330000; and pressure of 160 mm. of fluid. There was no pleocytosis on the sixteenth day, but the cerebrospinal fluid protein was 128 mg./100 cc. Agglutinin titers on the twenty-first day were 1:12,800 for L. pomona, 1:200 for L. icterohemorrhagiae and negative for L. canicola. On day 144 the titers were 1:200 for L. pomona and negative for L. icterohemorrhagiae and L. canicola.

Comment: The diagnosis of leptospirosis due to L. pomona was made on the basis of the high titer of agglutinins on the twenty-first day. This was the only patient with either pneumonitis or an abnormal colloidal gold curve. Another unusual feature was the disturbance of liver function tests noted on the twenty-sixth day, the significance of which is unknown.

CASE XI. A. S., a twenty-three year old white man from Eastland, Texas, was admitted to the Dallas Veterans Administration Hospital on February 2, 1949, on the sixth day of an illness characterized by onset with a chill, and by severe and continuous frontal headache, stiffness of the neck, generalized aching, fever and vomiting. There was malaise for one day preceding onset of illness. Sulfadiazine and penicillin therapy was begun soon after onset of illness and he felt better for two days, but his symptoms were very severe on the fifth day of illness, and on the morning of admission his temperature rose to 104°F. He had been employed as an animal caretaker for the Humane Society until eleven days prior to onset of illness.

The temperature was 99.6°F.; pulse, 84; and blood pressure, 125/80 mm. Hg. The eyelids were puffy, the conjunctivas were pink and the neck was slightly stiff. A scanty, erythematous, macular rash was noted on the trunk on the night of admission.

The hemoglobin was 14.4 gm./100 cc.; white blood cell count, 13,400/cu. mm.; and neutrophils, 79 per cent. The urine contained 1-plus protein and a few white cells and many red blood cells per high power field.

Myalgia and vomiting subsided on the day of admission but headache persisted and became more intense just before midnight of the seventh day then subsided twenty-four hours later. His temperature fell to normal soon after admission but after a few hours there was a second elevation which persisted for two days, with a peak of 102°F. on the eighth day. On the seventh day the rash was noted to involve the face, neck, trunk and extremities, with sparing of the palms and soles. In some areas it was confluent, and in a few areas it appeared urticarial. It persisted until the eleventh day. Chlortetracycline and penicillin therapy was started on the ninth day and continued for three days. Lumbar puncture on the seventh day revealed normal pressure, 12 cells/100 cc., and normal protein and sugar. No leptospiral agglutinins were detected on the seventh day, but on the eighteenth day the titers were 1:1000 for L. icterohemorrhagiae and 1:100 for L. canicola, the only antigens used.

Comment: The rise in agglutinin titers was sufficient evidence for the diagnosis of leptospirosis but the identity of the serotype remained in doubt because of the limited number of antigens used and the rather low titer of agglutinins for L. icterohemorrhagiae. Rash was a prominent feature in this case, and might possibly have been due to either sulfonamides or penicillin, but since similar rashes have been described so frequently in leptospirosis there is no need to invoke another explanation. The exacerbation of headache during the secondary rise in temperature probably represented mild clinical meningitis.

CASE XII.* G. B., a sixty-two year old white farmer from Sadler, Texas, entered the McKinney Veterans Administration Hospital on November 9, 1951, on the eighth day of an illness which began with a chill followed by myalgia, moderately severe frontal headache, malaise, anorexia and constipation. He was unaware of having fever but had experienced a feel-

* Previously reported [*11*].

ing of warmth below his waist. He noted orange-colored urine on the fourth day of illness, swelling of the legs on the fifth day, light stools on the seventh day and yellowness of the skin on the day of admission. There was no past history of hepatic, renal or cardiac disease, and no history of excessive intake of alcohol. He kept cows, pigs and horses on his farm and had noted rats around his barn, but there had been no contact with other rodents or with stagnant water.

The temperature was 100.4°F.; pulse, 84; and blood pressure, 148/90 mm. Hg. The skin and eyes were icteric and there was conjunctival suffusion, but no nuchal rigidity, pulmonary rales, cardiac enlargement or hepatosplenomegaly. There was pitting edema of the legs extending above the knees.

The white blood cell count was 24,400/cm. mm.; neutrophils, 92 per cent; hemoglobin, 12.4 gm./100 cc.; and sedimentation rate, 35 mm. (Wintrobe). The urine specific gravity was 1.016, there was 2-plus protein and the test for bile was positive. Subsequent urinalyses were all normal except for the presence of bile. The thymol turbidity was 13 units; cephalin-cholesterol flocculation test, 4 plus; serum alkaline phosphatase, 7.7 Bodansky units; bilirubin, 12.5 mg./100 cc.; albumin, 2.6 gm./100 cc.; globulin, 2.3 gm./100 cc.; cholesterol esters, 70 mg./100 cc.; and blood urea nitrogen, 25 mg./100 cc. The white blood cell count rose to 35,000/cu. mm. on the second hospital day, then fell progressively to normal, but the sedimentation rate was still 35 mm. after four weeks. All hepatic function tests gradually improved, and after five weeks the bromsulphalein retention was 4 per cent in forty-five minutes; serum albumin, 3.9 gm./100 cc.; thymol turbidity, 6 units; cephalin-cholesterol flocculation test, 1 plus; and bilirubin, 1.5 mg./100 cc.

The patient was relatively free of symptoms in the hospital even though he had low grade fever until the seventeenth day of illness. Lumbar puncture on the eleventh day disclosed xanthochromic cerebrospinal fluid with protein of 90 mg./100 cc. but no increase in cells. Diuresis began spontaneously on the thirteenth day and there was loss of 40 pounds of weight in the next ten days. Serial agglutination tests were positive for L. grippotyphosa in titers of 1:400 on the thirteenth day and 1:25,600 on the seventeenth day.

Comment: This case illustrates the occurrence of severe hepatic damage in the absence of other features of Weil's disease. The first stage was rather mild, but evidence of hepatic injury appeared by the fourth day and by the eighth day he had frank icterus, edema and abnormal liver function tests. The edema could be attributed only to hepatic damage since there was no evidence of significant renal damage or heart failure. Of incidental interest is the fact that this was the first case of L. grippotyphosa infection reported in the United States.

CLINICAL FEATURES

Stages of Illness. It is obvious from a review of the cases seen at this hospital that leptospirosis is a biphasic illness. Initially, there is a five- to seven-day period of fever, chills, headache, myalgia, conjunctivitis and gastrointestinal symptoms. Since leptospires can be recovered from the blood and cerebrospinal fluid during this phase in a high percentage of cases [*4,12*], it has been designated as the "septicemic" stage. The second stage begins as defervescence and symptomatic improvement occur on the sixth or seventh day. Shortly following the end of the first stage there is usually a secondary rise in temperature and development of meningitis, either asymptomatic or clinical. It has been demonstrated repeatedly that the blood and cerebrospinal fluid are sterile during this stage [*4,12*], organisms rarely having been recovered after the seventh or eighth day. During this second stage antibodies appear in the blood in rapidly rising titer [*4*]. In addition to fever and meningitis, several infrequent manifestations such as iridocyclitis [*4*] and various neurological lesions may develop [*12,13*]. It is my opinion that the second stage manifestations are primarily due to hypersensitivity, and it is proposed that this phase be designated as the "immune" stage. The previously suggested terms "icteric" stage [*14*] and "toxic" stage [*15*] are inappropriate because of the low incidence of jaundice in leptospirosis in general [*5*], and because there is little evidence that toxins are important in the production of the principal manifestations. Gsell [*4*] stressed the biphasic character of leptospirosis in his monograph.

Manifestations. The various manifestations observed in this study are listed in Table I, and for comparison those observed by McCrumb et al. [*5*] in their large series are recorded in Table II.

First Stage. In the majority of cases the onset of illness was abrupt, but in Case III it was insidious, and in Case XI malaise preceded the onset of other symptoms by twenty-four hours. Headache and myalgia were the outstanding symptoms in most of the cases. The headache was usually frontal in location (sometimes bitemporal or occipital), and was described as intense and unremitting (rarely throbbing) in character. It tended to persist throughout the first stage. That it was not a manifestation of meningitis was demonstrated by lumbar punc-

TABLE I
INCIDENCE OF CLINICAL MANIFESTATIONS IN TWELVE CASES OF LEPTOSPIROSIS

Manifestation	Cases (no.)	Manifestation	Cases (no.)
Fever	12	Retrobulbar pain	2
Chills	12	Rash	2
Headache	12	Photophobia	2
Meningitis	10	Diarrhea	1
Myalgia	9	Pneumonitis	1
Conjunctivitis	9	Jaundice	1
Anorexia	9	Edema	1
Nausea and vomiting	5	Palpable spleen	1
Nuchal rigidity	5	Palpable liver	1
Abdominal pain	4	Delirium	1

TABLE II
CLINICAL MANIFESTATIONS OF LEPTOSPIROSIS (MCCRUMB ET AL. [5])

Symptom-Sign	Military Patients			Civilian Patients		
	Total Recorded	No. Positive	%	Total Recorded	No. Positive	%
Headache	172	164	95.3	28	26	93
Gastrointestinal symptoms	168	161	95.8	28	27	96.4
Conjunctival injection	169	144	85.2	28	23	82.1
Myalgia	173	134	77.4	28	16	57.1
Respiratory symptoms	165	117	70.9	29	25	86.2
Abrupt onset	158	111	70.2	24	24	100
Retrobulbar pain	154	85	55.1	27	11	40.8
Lymphadenopathy	166	74	44.5	29	15	51.7
Muscle tenderness	51	22	43	24	16	66.7
Abdominal tenderness	59	25	42.3	24	13	54.1
Nuchal rigidity	163	59	36.2	30	11	36.6
Photophobia	153	47	30.7	27	7	25.9
Joint pain	155	45	29	28	18	64.2
Hemorrhagic signs	160	44	27.5	29	7	24.1
Rash	164	44	26.8	28	5	17.8
Splenomegaly	162	26	16	29	5	17.2
Hepatomegaly	160	22	13.7	30	5	16.6
Jaundice	150	6	3.8	27	5	18.4

ture in three patients who were found to have a stiff neck on admission. Muscular discomfort was maximal in the calf and lumbar muscles, but other muscle groups were affected as well. There was muscle tenderness only in the patients having severe myalgia. Abdominal pain, such as that seen in four patients, has been attributed to involvement of muscles rather than of the gastrointestinal tract [*3*].

Conjunctivitis was not observed in all patients but it proved to be a valuable diagnostic clue when present. In Case IX the eyes were so red that the patient himself had noted their color, but in others the congestion was barely detectable. Usually only the bulbar conjunctivas were suffused, giving the eyes a bright pink appearance similar to that which occurrs after the eyes have been exposed to fresh water during swimming. There was no exudate or edema but one patient had puffy eyelids. Burning, photophobia and retrobulbar pain were occasional complaints.

All the patients had fever and chills. The fever was pronounced, with the temperature often exceeding 104°F., and chills were recurrent in the majority of cases. Some patients experienced chills on as many as five different days. A number of patients were given antibiotics during the first stage but there was no detectable alteration in the degree or duration of fever.

The majority of patients had minor gastrointestinal complaints such as anorexia and constipation, and in one instance, mild diarrhea. Vomiting was the only important gastrointestinal manifestation noted, however. Hematemesis and melena have been described [*16*] but were not observed in this study. Although abdominal pain is considered to be the result of muscular involvement, its occurrence in patients who are vomiting and have abdominal rigidity and tenderness may lead the physician to suspect acute appendicitis or cholecystitis, and a few such patients have been subjected to surgery [*3*].

Rash occurred in two patients. In Case II it was a blotchy, erythematous eruption which disappeared overnight, but in Case XI there was an extensive maculopapular involvement of the trunk, neck, face and extremities which persisted for five days. In some areas the lesions were confluent, in others urticarial. One patient had cough and roentgenographic evidence of pneumonitis, the only significant pulmonary involvement. Arthralgia was a major complaint in one case. Only one patient had jaundice which, incidentally, has been regarded by others [*2,15*] as a second stage manifestation even though there is good evidence that hepatic damage occurs during the septicemic stage, as in Case XII. In this case there were none of the other manifestations of Weil's disease and, except for severe hepatic damage, it would have been one of the mildest cases of the series.

The routine laboratory observations disclosed a range of leukocyte counts from 4,400 to 13,400/cu. mm. in the anicteric patients, while the jaundiced patient had marked leukocytosis,

one count of 35,000/cu. mm. being recorded. Neutrophilia was a constant finding, as was persistent elevation of the sedimentation rate. Proteinuria and cylindruria were detected in two-thirds of the patients seen in the first stage but the urine rapidly cleared after defervescence. None of the patients with proteinuria had evidence of significant impairment of renal function, but one patient (Case VI) had elevated blood urea nitrogen in the presence of normal urine. The anicteric patients had only minor disturbances of liver function tests, but in Case XII the patient had both clinical and laboratory evidence of severe hepatic damage.

Second Stage. The second stage in the anicteric patients was remarkably benign. Whereas patients with Weil's disease usually remain ill for weeks (often critically so) [*16*], none of these patients were seriously ill in the second stage. Several, including some who had overt meningitis, were clinically well by the tenth or eleventh day of illness. The two patients with delayed onset of meningitis remained symptomatic for approximately four and three weeks, respectively, but the others were all asymptomatic by the end of the second week. The jaundiced patient, on the other hand, had slow recovery and was febrile until the seventeenth day and did not complete diuresis until the twenty-third day. The prognosis in anicteric leptospirosis is universally good, and residual damage in any type of leptospirosis is rare [*2*].

The clinical picture in the second stage is entirely different from that in the first stage. Fever is common, but its duration is usually brief, two or three days, and the temperature rarely exceeds 102°F. In several cases (Cases I, III, V and X) low grade fever persisted after the first stage without a break, while in others (Cases II, IV, VI, IX and XI) there was an afebrile interval of twelve to thirty-six hours preceding its onset. This latter sequence of events resulted in the biphasic temperature curve observed so frequently by Gsell [*4*] in L. pomona infections. However, it is not peculiar to any particular serotype, nor even to leptospirosis, as the same pattern is seen in dengue [*17*].

Since the blood and cerebrospinal fluid are sterile in the second stage, the manifestations must have a different pathogenesis than those of the first stage. The fever of the second stage occurs at a time when antibody titers are rising rapidly and leptospires are being destroyed, thus suggesting the possibility that it is on an allergic basis. Febrile relapses of the type observed in Cases VII and VIII may be on the same basis, as suggested by Inada [*14*], who regarded such febrile episodes not as relapses but as "after fever." He considered the fever to be an allergic reaction resulting from the absorption of leptospiral products from the kidneys at the height of immunity (about the fourteenth day). Meningitis, the principal second stage manifestation, developed in ten of eleven patients in whom cerebrospinal fluids were examined. This feature is discussed in detail in the next section. None of the rare second stage manifestations, such as iridocyclitis, peripheral neuritis, myelitis, alopecia and leptospiral endocarditis, have been seen at this hospital.

Meningitis. The incidence of meningitis in this small series was comparable to that reported by Gsell [*4*], Minkenhoff [*18*] and Cargill [*20*]. Not all the patients had overt meningitis, however, since three patients had pleocytosis without any of the usual signs or symptoms of meningitis, while another patient (Case I) had severe delirium beginning on the sixth day and, although he had pleocytosis, he was not considered to have clinical meningitis.

The usual sequence of events is for leptospiral meningitis to make its appearance some twelve to twenty-four hours after the first stage defervescence; however, in two cases the onset was delayed until the end of the second week. Gsell [*7,8*] has described two such patients also. The onset of clinical meningitis was heralded in every case by sudden recurrence of headache or by an intensification of headache if that of the first stage had not subsided. The headache associated with meningitis was even more distressing than that of the first stage, frequently requiring the administration of codeine or meperidine for relief. Any exacerbation or recurrence of headache on or after the sixth day should be regarded as a manifestation of meningitis. Lumbar puncture afforded striking relief of headache in some of the patients with meningitis.

The three patients (Cases I, III and IV) with nuchal rigidity and normal spinal fluid illustrate the fact that stiffness of the neck in the first stage of leptospirosis does not indicate meningitis. Moreover, in the second stage nuchal rigidity may be absent, even in the presence of symptomatic meningitis (Case V). It should be remembered that meningitis cannot be excluded on clinical grounds alone, or on the basis of lumbar puncture performed before the seventh

TABLE III
CEREBROSPINAL FLUID OBSERVATIONS

Patient	Case	Day of Illness	White Blood Cells (cu. mm.)	Polymorpho-nuclears (%)	Lymphocytes (%)	Protein (mg. %)	Sugar (mg. %)
L. L.	I	4	0	..	...	34	89
		6	11	66	44	30	73
C. B.	III	5	0	..	...	17	81
		7	288	25	75	140	100
		12	95	2	98	33	73
		31	112	2	98	56	65
		60	12	8	92	36	...
J. C.	IV	6	0	..	...	41	85
		12	35	11	89	17	...
		50	10	..	100	39	...
J. P.	V	7	382	91	9	102	73
		14	17	18	82	53	...
O. D.	VI	11	180	66	34	66	59
		41	4	..	100	39	...
M. V.	VII	23	533	2	98	91	65
		26	450	9	91	89	...
		30	287	3	97	61	...
		35	102	2	98	46	...
		46	42	..	100	41	...
		74	9	..	100	48	...
R. M.	VIII	13	406	..	98	113	69
		16	376	5	95	70	69
		22	133	30	70	47	...
		35	18	11	89	35	...
C. R.	IX	8	13	15	85	45	89
B. G.	X	10	56	76	24	82	63
		16	0	..	...	128	49
A. M.	XI	7	12	..	100	34	77
G. B.	XII	11	0	..	...	90	100

day [*19*]. Meningitis later developed in three patients in whom cerebrospinal fluids were normal at the time of the initial examination.

There was great variation in the severity of meningitis in this series. Three patients were asymptomatic, five had comparatively mild symptomatic meningitis, while two had severe meningitic syndromes with headache, vomiting and nuchal rigidity. There was also great variation in the duration of symptoms, from one day (Case III) to thirteen days (Case VI). Pleocytosis persisted long after subsidence of symptoms, until the sixtieth day in Case III and the seventy-fourth day in Case VII, but complete recovery occurred in every case.

The cerebrospinal fluid observations are recorded in Table III. The maximum degree of pleocytosis was found on the seventh day or on the first examination thereafter (except in Case I), and the range of maximum cell counts was 11 to 533/cu. mm. These values are comparable to those usually given [*20,21*], but counts of several thousand are on record [*4,20*]. On the initial examination, the predominant cell type was the neutrophil in four cases and the lymphocyte in six cases, but lymphocytes predominated after the twelfth day. The protein contents varied from normal to 140 mg./100 cc., and the sugar was normal or slightly elevated. The patient with jaundice had xanthochromic fluid.

Relatively little has been written regarding the pathogenesis and significance of meningitis in leptospirosis. Walch-Sorgdrager [*22*] suggested the possibility of increased cells and protein being produced by certain chemical substances rather than by the presence of leptospires in the meninges. Rosenberg [*6*] attributed the meningitis to invasion of the meninges by leptospires, but this is not a tenable explanation in view of the numerous reports of the isolation of leptospires from otherwise normal fluid. There appears to be little reaction to the mere presence

of leptospires in the meninges. Moreover, the organisms rapidly disappear from the cerebrospinal fluid following the onset of meningitis [*4*].

I am in agreement with the view of Gsell [*4*] that leptospiral meningitis is the expression of an antigen-antibody reaction, rather than a manifestation of direct injury to the meninges by leptospires. There have been a number of demonstrations that similar meningeal reactions can be elicited by the injection of bacteria or their products into the subarachnoid space of subjects exhibiting hypersensitivity to the bacteria. Burns and Finley [*23*] found no clinical or pathological response when tuberculin or tubercle bacilli were injected into the subarachnoid space of non-tuberculous animals but the same procedures caused an outpouring of cells and protein in hypersensitive animals. Cairns, Smith and Vollum [*24*] demonstrated similar reactions to the instillation of old tuberculin into the subarachnoid space of tuberculin-positive patients, and responses of the same type have been reported following intrathecal injection of streptococcal antigens [*25*]. In leptospirosis the meningeal reaction occurs only after there has been time for hypersensitivity to develop.

This theory explains the virtual absence of pleocytosis in the first stage, the abrupt onset of meningitis around the seventh day, the rapid disappearance of leptospires from the cerebrospinal fluid after onset of meningitis, the good prognosis, and the lack of correlation between severity of meningitis and manifestations of virulence. The high incidence of meningitis is explained by the remarkable ability of leptospires to enter the cerebrospinal fluid during the septicemic phase.

LABORATORY DIAGNOSIS

McCrumb et al. [*5*] required satisfaction of one or more of the following criteria for the laboratory diagnosis of leptospirosis: (1) isolation of leptospires from the blood; (2) fourfold or greater rise in complement-fixing or agglutination-lysis antibody titers, or both, during the course of illness; (3) sustained titer of 1:40 or greater by complement fixation, or 1:400 or greater by agglutination-lysis when the first serum specimen was obtained on or after the seventh day of illness.

All cases in this series, with one exception, met one or more of these criteria. The diagnosis of leptospirosis was made in Case x on the basis of a high titer of agglutinins, 1:12,800, on the twenty-first day and the persistence of a titer of 1:200 after 144 days. Serums in all cases were tested by the microscopic agglutination-lysis technic using live antigens. Serums in Case xii were tested against sixteen antigens, serums in Cases i to viii against nine antigens (L. icterohemorrhagiae, L. canicola, L. pomona, L. grippotyphosa, L. hebdomadis, L. mitis, L. autumnalis A, L. autumnalis AB and L. bataviae), serums from Cases ix and x against three antigens (L. icterohemorrhagiae, L. canicola and L. pomona) and serums in Case xi against two antigens (L. icterohemorrhagiae and L. canicola). There was no attempt to isolate leptospires except in Cases i and ii. In Case ii, L. pomona was cultured from blood drawn on the fourth day. It is obvious that a precise identification of the infecting serotype by serological tests is often impossible.

COMMENTS

The clinical experience at this hospital does not support the view that different leptospiral serotypes cause distinctive clinical patterns of disease [*26*]. It is impossible to make a species diagnosis on the basis of clinical grounds alone in any of these cases. Clinically, Cases vii and viii were practically identical, but the infecting serotypes were different. Moreover, one patient (Case viii) had "aseptic meningitis" and the other (Case xii) had severe hepatic damage without meningitis, yet the etiological agent in each case was L. grippotyphosa. There are no clinical syndromes exclusively linked with infection by specific serotypes, nor are there any serotypes which always cause the same pattern of disease. This observation has been confirmed by the extensive experience of McCrumb [*5*] in Malaya, and Van Thiele [*16*] and Borg-Petersen [*27*] in Europe.

Names such as "mud fever," "swamp fever" and "swineherd's disease" should be used only to draw attention to the epidemiological features of a given case, not to the identity of the serotype; and use of the terms "Weil's disease" and "pretibial fever" should be limited to the description of certain syndromes rather than as synonyms for infection with L. icterohemorrhagiae and L. autumnalis, respectively. When special names are used the causal serotype should always be indicated if possible.

All the features of leptospirosis can be explained on the basis of the biphasic concept of

the disease. There is general agreement that the clinical manifestations of the first stage are directly related to leptospiremia, but the role of hypersensitivity in the pathogenesis of the second stage manifestations has not been generally appreciated, even though it has been mentioned by several authors. Gsell [*4*] regarded meningitis as the expression of an antigen-antibody reaction, Middleton [*28*] proposed a hypersensitivity reaction to account for the rare neurological lesions, and Inada [*14*] and Davidson [*29*] considered "after fever" to be an allergic phenomenon. The elevation of temperature occurring early in the second stage at the time when antibody titers are rising and leptospires are being destroyed is probably on an allergic basis also. On the other hand, there are two manifestations, leptospiruria and iridocyclitis, which result from the proliferation of leptospires in organs in which the concentration of antibodies remains very low, the kidneys [*29*] and the anterior chamber of the eye [*30*], respectively.

There is every reason to believe that leptospirosis is widespread in the United States, just as it is throughout the world. No doubt leptospirosis is present in many patients and is now being called "summer influenza," "aseptic meningitis" and "non-paralytic polio." Among inhabitants of villages and rural areas leptospirosis is predominant in the summer and autumn, but among urban residents the seasonal incidence is less marked. As physicians become increasingly aware of the milder leptospiral infections, and with improvement in laboratory facilities for diagnosis, the number of cases being reported to public health agencies should be greatly increased.

SUMMARY

A study of twelve sporadic cases revealed leptospirosis to be an acute, self-limited, diphasic illness with a low incidence of jaundice or significant renal damage. The first stage manifestations were compatible with the septicemic state which is known to occur during the first five to seven days, but the clinical picture in the second stage appeared to be the consequence of the body's immunologic response. It is proposed that this phase be referred to as the "immune" stage rather than as the "icteric" or "toxic" stage.

It was impossible to correlate the different clinical disease patterns with the leptospiral serotypes identified in these cases. For this reason the use of special names as synonyms for infections with certain species is not justified.

The diagnosis of leptospirosis should be considered in any patient with an acute illness characterized by headache, myalgia, fever, chills and conjunctival suffusion, and in any patient in whom "aseptic meningitis" develops following an acute febrile illness of five to seven days' duration.

The greatest danger of infection in man is from contact with stagnant water to which wild or domesticated animals have access, but infection may result from direct contact with the urine of infected animals as well.

Acknowledgment: I am indebted to Dr. Donald Seldin, the University of Texas Southwestern Medical School, for his suggestions and constructive criticism; to Dr. Robert Pike, the University of Texas Southwestern Medical School, for performing serological tests in the first eight cases; and to the Veterinarian Division of the Walter Reed Army Institute of Research, Washington, D. C., for performing serological tests in the last four cases.

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Asian Influenzal Pneumonitis*

A Structural and Virologic Analysis

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RECENT outbreaks of Asian influenza were first reported in the latter part of February 1957 from the Chinese province of Kweichow [*1*] and later in April 1957 from Hong Kong and Singapore. In rapid succession, epidemics occurred in the Philippines, the Malayan States, Japan, India and Russia. The Asian influenza virus apparently reached the United States in June 1957, indicated by the simultaneous outbreaks of influenza epidemics on the eastern and western coasts [*2*]. From these coastal regions the virus was carried inland to the central states and was noted in the greater St. Louis area in the first week of September 1957 by the virologic identification of an influenza epidemic in a St. Louis county school. Similar occurrences were reported in the general vicinity for the next eight weeks; but by the end of November, the epidemic was rapidly subsiding.

The lethal effect of the virus was attested to by reports from general hospitals in the St. Louis area which indicated an appreciable increase in mortality from influenza and pneumonia over the previous year. The combined deaths from influenza and pneumonia, in the St. Louis University group of hospitals, numbered nine in the period from September 1 to December 1, 1956. During the same period in 1957, forty-nine fatalities were attributed to these causes [*3*].

MATERIALS AND METHODS

In all instances the presenting symptoms in the nine fatal cases, which were confirmed by virologic or postmortem studies, were those of the "flu syndrome" characterized by an abrupt onset of fever, sore throat, cough, chills and myalgia. Of the patients discussed in this paper, rapid deterioration of their condition occurred from the onset of symptoms with death being due to respiratory or cardiorespiratory insufficiency despite supportive measures including the administration of antibiotics, cardiac glycosides, oxygen, intermittent positive pressure breathing and adrenal cortical steroids.

Of the nine deaths attributed to Asian influenzal pneumonitis, six were proved by virologic studies. Of these fatal cases five were in females and four in males, with the patient's age varying from twenty-one to fifty-four years. The duration of illness from the onset of symptoms to death ranged from two to fifteen days. None of these patients had been vaccinated against the Asian strain of the influenza virus. Another significant feature in all but three of these cases was the existence of an antecedent physiologic or an established pathologic state. Of the five women, four were in the later stages of pregnancy or had recently delivered. (Table I.)

Of the six patients from whom the virus was recovered, bacteria were cultured from the lungs of four; Staphylococcus aureus was recovered from two, anaerobic streptococci from one and diphtheroids were grown in two patients. Cultural recovery of the diphtheroids probably represents postmortem contamination, as these organisms do not usually possess pathologic significance. The serum in one case showed a heterophil titer of 1:1280 and after absorption with guinea pig kidney, the titer was 1:448, but after beef cell absorption studies negative results were obtained. These findings and the clinical evidence of enlarged lymph nodes prior to delivery of a live infant, suggested an antecedent or coexistent case of infectious mononucleosis in the mother. In the seven cases of Asian influenza in which blood cultures were obtained no bacterial growth was found. In the three cases in which the virus was not recovered, postmortem procedures precluded the possibility of performing specific diagnostic technics; a presumptive diagnosis of viral pneumonitis was based on characteristic histopathologic features.

Isolation of the Asian influenzal virus was accomplished by the inoculation of embryonated eggs with prepared lung tissue secured under aseptic technic at autopsy. At the same time lung tissue was also submitted for bacteriologic cultures. Serum specimens were obtained from three patients (Cases 2, 4 and 8) antemortem. The antihemagglutin titer against

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TABLE I
SUMMARY OF CLINICAL FINDINGS

Case (No.)	Patient's Initials	Age (yr.)	Sex	Duration of Illness	Associated Conditions	Findings on X-rays of Lungs
I	R. G.	35	M	48 hr.	None	None
II	S. M.	54	M	90 hr.	Severe coronary atherosclerosis and old myocardial infarction	Diffuse bilateral involvement
III	I. R.	40	F	96 hr.	None	None
IV	M. B.	33	F	7 days	Mitral stenosis; Type II nephritis; uterine pregnancy, 8 mo., 2 days postpartum	Involvement of lower left lobe
V	R. A.	21	F	7 days	Mitral stenosis; uterine pregnancy, 7 mo.	Diffuse involvement of right lung
VI	S. G.	30	F	7 days	Uterine pregnancy, 26 wk.	Diffuse bilateral involvement.
VII	A. H.	42	M	11 days	None	Diffuse involvement of right lung
VIII	G. T.	22	F	12 days	Uterine pregnancy, 7 days postpartum; infectious mononucleosis, suspected	Diffuse bilateral involvement
IX	R. G.	51	M	15 days	Fiedler's myocarditis; Laennec's cirrhosis	Diffuse involvement of right lung

Asian influenza virus JAP-305-57 was quite high in the latter two cases and was rising in Case 2. (Table II.)

The whole lung sections (Figs. 1, 2 and 3), were prepared using the macro method technique with complete formalin insufflation of the pulmonary tissue via the tracheobronchial tree. After fixation, the lungs were embedded in 100 bloom gelatin, frozen, and cut at a thickness of 100 microns, mounted on paper and photographed on a transilluminator. In several cases, the lung microscopic alterations were studied by ultra soft x-ray to elicit information concerning the density of the hyaline membrane. Histopathologic examination of the other viscera was by conventional technics.

PATHOLOGY

The lungs in each case exhibited distinctive lesions on both gross and microscopic examination, more marked in some examples than others. In those lungs in which there was superimposed bacterial infection, an attempt was made to

TABLE II
SUMMARY OF VIROLOGIC AND BACTERIAL STUDIES

Case (No.)	Patient's Initials	Virus Isolation from Lungs	Antibodies against Jap 305-57	Bacterial Isolation	
				Lung	Blood
I	R. G.	I	NA	Anerobic, streptococci and diphtheroids	N
II	S. M.	I	1:40	Staphylococcus aureus	N
III	I. R.	NA	NA	NA	NA
IV	M. B.	I	1:1280	Staphylococcus aureus	N
V	R. A.	I	NA	NA	N
VI	S. G.	I	NA	NA	N
VII	A. H.	NA	NA	NA	N
VIII	G. T.	I	1:320	Diphtheroids	N
IX	R. G.	NA	NA	NA	NA

NOTE: I—isolated.
N—not isolated.
NA—isolation not attempted.

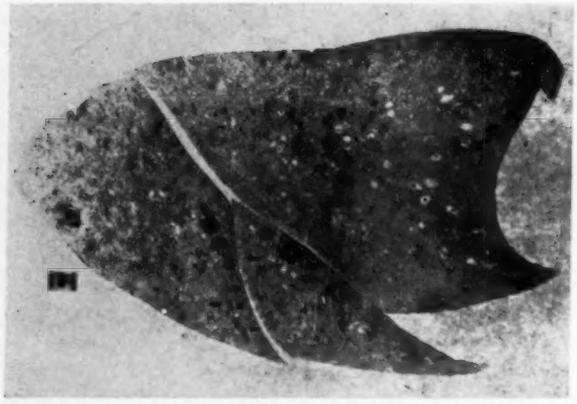

FIG. 1. Case IV. Whole lung section showing focal, basilar and septal distribution of the hemorrhagic areas.

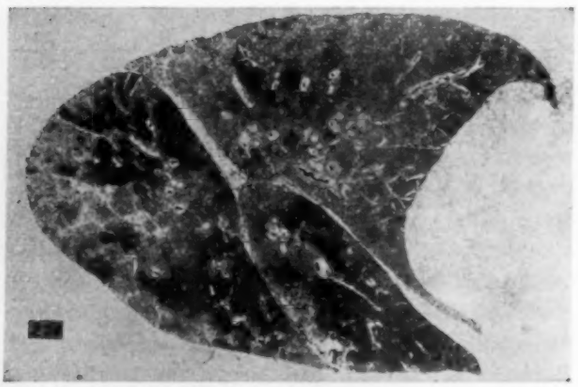

FIG. 2. Case VI. Whole lung section showing the extensive lobular pattern of the hemorrhagic areas. All macrosections 100 microns thick and unstained.

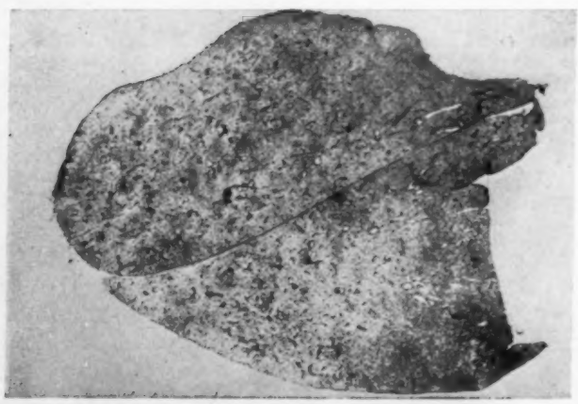

FIG. 3. Whole lung section of normal lung.

delineate the changes due to the virus and those due to bacteria.

The severest macroscopic alterations noted in most instances were predominantly lobular in situation, but in Case 6 the lesions were lobar in distribution. The affected pulmonary areas varied in color from deep scarlet to purple and in consistency from subcrepitance to firmness. The cut surface of all lungs oozed red, frothy fluid and in Cases 4 and 6 gross hemorrhage was present. The distribution of the hemorrhagic areas is demonstrated in both whole lung sections. (Figs. 1 and 2.) The large lung sections depicting Asian influenza pneumonia compared with the normal lung (Fig. 3) showed the dark areas due to hemorrhage with a background of diffuse reticular involvement.

The bronchi and grossly visible bronchioles contained red, frothy fluid in most cases indicative of a tracheobronchitis. The hilar lymph nodes were succulent and enlarged. Pleural effusion was not a constant finding and was limited in amount up to 300 cc. in the cases in which it was present.

The histopathological changes are discussed in a composite manner, within a temporal evolutionary framework. The histopathologic reaction was characterized essentially by a mononuclear cell response which was predominantly interstitial in location. The alveolar septums were thickened by edema fluid, capillary congestion and cellular elements of the lymphocytic and mononuclear types. The alveolar spaces were clear in some areas, but in the regions of marked structural alterations eosinophilic edema fluid and macrophages were prominently displayed. In small focal areas the alveoli contained neutrophilic leukocytes, cellular debris, and a spun fibrin network. The latter changes were best seen in Cases 3 and 5. In Case 7 numerous small abscesses were noted in addition to the changes already mentioned. In all instances, the inner surface of many of the alveoli was covered with an eosinophilic band-like hyaline membrane. (Fig. 4.) Underlying this membrane, the septal walls were thickened and often lined by cuboidal epithelium. Frank necrosis of the alveolar walls with hemorrhage into the alveolar lumens and capillary thrombosis were found in most cases. These latter changes were marked in Cases 3, 5 and 6. Stains such as periodic-acid-Schiff, Alcian blue, and Goodpasture's gram stain indicated that the tinctorial reaction given by this intra-alveolar and intraductal membrane

was akin to that seen in other conditions in which eosinophilic hyaline membranes have been described, such as Type A influenza or other viral pneumonias. Figure 5 is a soft x-ray microradiograph demonstrating the remarkable density of the hyaline membrane, in sharp contrast to the congested septal walls. To the best of our knowledge, this represents the first time that ultra soft x-rays have been used to qualitatively analyze this intra-alveolar membrane. The hyaline membrane even in early cases was closely apposed to the alveolar wall or ducts. An associated alteration of considerable interest was the presence of sudanophilic vacuoles within the membrane resulting in its fragmentation and disruption. This lipid material was probably derived from the serum, having escaped through the injured capillaries. (Fig. 6.) Recent work suggests that the basic constituent of the membrane is globulin derived from the serum. The presence of the lipid complex associated with the PAS positive hyaline would support the intravascular origin of the hyaline membrane. In some areas, a giant cell reaction was noted surrounding this vacuolated material. (Fig. 7.) In cases of three to seven days standing, the membrane showed a laminated pattern and the interlaminar cement material took a positive gram stain for fibrin. This lamellar pattern suggests a gradual buildup of hyaline material over a period of several days, with the thread of fibrin binding the hyaline material. Another cardinal feature was the overdistention of the alveolar ducts. This overdistention is probably due to two factors: a partial block in the terminal bronchioles by exudate and edema and a partial interference with collateral air drift, as the hyaline membrane temporarily obstructs the pores of Cohn or Lambert's channels. (Fig. 8.)

Bronchi and bronchioles showed marked congestion and mononuclear cell infiltration in the subepithelial zone; in other areas exfoliation of the epithelium was prominent. In Case 6, frank ulceration of the lining of the terminal bronchiole was noted with purulent material in the lumen. Pleuritis was an unknown feature in these cases. (Table III.)

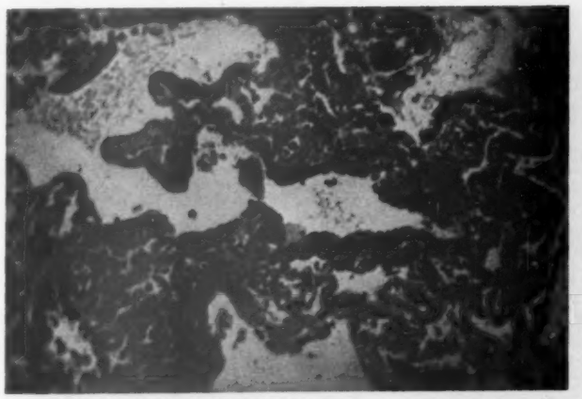

FIG. 4. Marked interstitial inflammation of septal walls; hyaline membrane outlining dilated alveolar ducts. Hematoxylin and eosin, original magnification × 180.

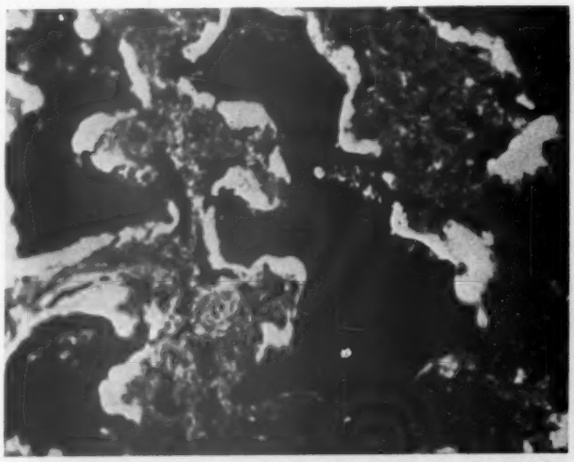

FIG. 5. Microradiograph demonstrating the density of the hyaline membrane which is intimately associated with alveolar walls. Original magnification × 180. 2.5 KV; 3.5 m. Amp.

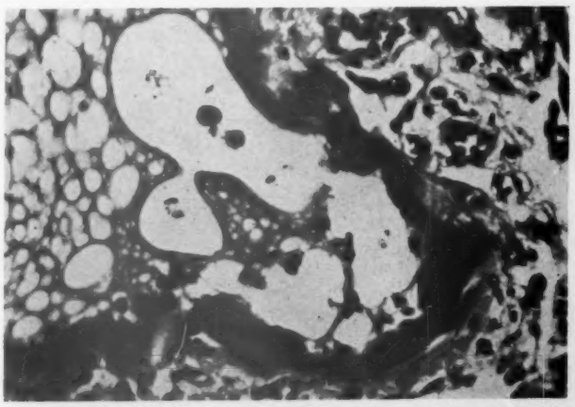

FIG. 6. Sudanophilic vacuoles incorporated with hyaline membrane; the latter illustrating a laminated pattern. Hematoxylin and eosin, original magnification × 320.

COMMENTS

The establishment of a correct diagnosis in an epidemic is of the greatest importance and numerous means are available to accomplish this. Included in these means are serologic, virologic and bacteriologic studies, both pre-

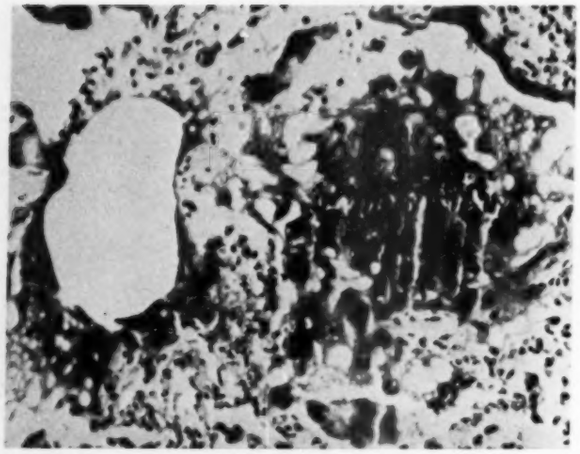

FIG. 7. Foreign body reaction to sudanophilic globules and fragmented hyaline membrane. Original magnification × 320

and postmortem, as well as necropsy examination. If a necropsy cannot be performed, one technic of interest and easily performed is to obtain a needle biopsy specimen of the lungs immediately after death to secure tissue for cultures and microscopic study. Such a technic should be more widely used in establishing a diagnosis in any future respiratory disease epidemics such as the recent one of Asian influenza. The material garnered contributes variable information on the specificity of lesions, the extent of the disease in the general population, and possibly the existence of the virus in the lungs without the existence of histopathologic alterations, from individuals dying from some other cause.

Interestingly enough, several infants who died during this same influenzal epidemic and whose lungs revealed marked interstitial pneumonia were free of hyaline membrane formation and also of the Asian strain of influenza virus. These sudden deaths in infancy are seen in the fall and winter months and are usually examined on the medicolegal service. The inability to recover the influenza virus from the lungs in these cases enhances the significance of recovery in the fatal influenzal cases, indicating the virus is pathogenic and not universally present in the lungs of the general population during an epidemic without producing pulmonary lesions.

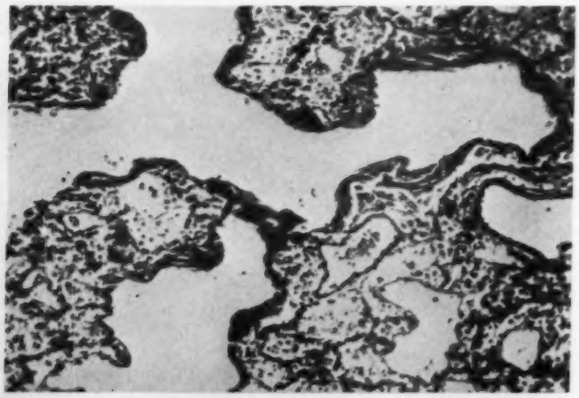

FIG. 8. Dilated alveolar duct of respiratory lobule lined by hyaline membrane. Hematoxylin and eosin, original magnification × 120.

With each succeeding case in this outbreak of Asian influenza it became apparent that the fatalities occurred early in the course of the disease and in relatively young persons. Another observation of importance was that the majority of fatalities developed on a background of antecedent disease or a physiologic state of pregnancy.

The cause and mechanisms of death in influenza epidemics have long evoked interest. In the influenza epidemics of 1889–1891 and the pandemics of 1918–1919 the paramount problem was the relationship between the superimposed bacterial infection and the initial viral infection. Even today this relationship has not been clearly defined; but the justified reliance on the efficacy of antimicrobial agents in suppressing bacterial infection allows a clear reappraisal of the nature and lethality of the viral infection. The potential eradication of bacterial infection and associated effects probably allows a more penetrating evaluation of the effects attributable to the action of the virus.

Bacterial and Viral Infection. In Wolbach's analysis of patients dying from influenza in 1918, the most frequent secondary invader present was Hemophilus influenza, although in some series the principal invader was Staph. aureus [*4*]. It is apparent that in fatal cases in more recent epidemics of influenza the most frequently recovered organism was Staph. aureus. In the 1949 influenza epidemic in Rotterdam, thirty-seven cases of pneumonia were observed, seventeen of which were caused by staphylococcus. Of these seventeen cases, fifteen were cases of influenza of the A′ type [*5*]. This relationship between influenza virus and staphylococcal infection was also emphasized in the Asian influenza epidemic in Chile in July 1957. Although various organisms were associated with the fatal cases of pneumonia in that epidemic, Staph. aureus was most frequently isolated [*6*].

TABLE III
SUMMARY OF HISTOLOGIC LESIONS

Case (No.)	Patient's Initials	Acute Bronchiolitis	Hyaline Membrane Formation	Alveolar Duct Emphysema	Exfoliation of Bronchial Epithelium	Alveolar Wall Necrosis	Bacterial Super-infection	Septal Repair
I	R. G.	A	P	P	P	P	P	A
II	S. M.	P	P	P	P	P	A	P
III	I. R.	P	P	P	P	P	P	P
IV	M. B.	P	P	P	P	P	P	P
V	R. A.	P	P	P	P	P	P	P
VI	S. G.	P	P	P	P	P	P	A
VII	A. H.	P	P	P	P	P	P	A
VIII	G. T.	P	P	P	P	P	A	P
IX	R. G.	P	P	P	P	A	P	A

NOTE: A—absent.
P—present.

In our series bacterial inflammation was noted microscopically in seven of nine cases. In these seven cases, lung tissue from only two patients was submitted for culture. These revealed streptococcus and diptheroids in one case and Staph. aureus in the other. Bacteria were cultured from the lungs of the remaining two patients (Cases 2 and 8) although there was no histologic evidence of their effects. The presence of bacteria in the lungs as demonstrated by culture and/or gram stain does not constitute valid evidence of a progressive bacterial infection. The essential criterion necessary for the diagnosis of the latter is the presumptive microscopic evidence of bacterial infection. It is reasonable then to conclude that these two cases of fulminant pneumonia were solely due to the influenza virus. The recent report of a fatal case of Asian influenzal pneumonia [7] in which no bacteria were isolated or seen, and in which there was no microscopic evidence of bacterial infection, has also been offered as documentation of the entity of pure influenzal pneumonia. In this instance, as well as those in our series, the same pathologic changes were seen as were noted during the 1918 pandemic and this suggests that the role of secondary bacterial invaders contributing to death may have been overemphasized.

On the question of transplacental passage of the influenza virus, a complete discussion is not possible, owing to the paucity of material available, as virologic and histopathologic investigation were only carried out in Case 4 (a baby). This baby died on the second day of life and at autopsy the lungs failed to reveal evidence of viral pneumonia or histologic or cultural grounds. The antihemagglutinin titer of the infant was less than 1:10. But, from these findings, it is inferred that neither the influenza virus nor maternal antibodies crossed the placenta.

Vascular Dynamics. Six cases were associated with the state of pregnancy, mitral stenosis, myocarditis, myocardial infarction and Laennec's cirrhosis. It is possible that these conditions have as a pivotal feature a functional alteration in vascular hemodynamics; and the structural alterations in the pulmonary vascular bed and air sac resulted in further embarrassment of an already impaired circulatory pathway. Two of the four pregnant women had mitral stenosis, although prior to pregnancy this valvular disability had caused little limitation of activity. As pregnancy advances there is a functional activation of mechanisms to increase cardiac output. This is accomplished by increasing the speed of circulation as well as the blood volume, both of which are tolerated by the normal heart. With a valvular disease of the heart an adequate output may not be possible in the face of these compensatory responses, the obvious consequence being cardiac insufficiency with attendant changes in the pulmonary vascular bed. These adaptative changes of the vascular system are most prominent between the fourth and eighth months of gestation and consequently this is the period of greatest concern [8]. Three

of the four deceased pregnant women were in this latter period and one had recently delivered. A rational explanation can be offered for the death of the women with mitral stenosis and pneumonia, but the disturbance of circulatory dynamics in the pregnant women with the normal hearts at autopsy is more difficult to ascertain.

Viral myocarditis is often unsuspected in many viral infections, and without histologic evidence of interstitial inflammation, the subtle structural changes within the myocardial fibers may be overlooked by conventional microscopic technics. A granulomatous myocarditis, recalling the so-called Fiedler's myocarditis, was present in Case 9.

In contrast to the antecedent conditions contributing to death in these cases of Asian influenza, Wolbach's investigation of the fatal influenzal cases at Camp Devens in 1918, does not emphasize or uncover underlying disease processes which may have been contributory factors in the death of young individuals.

Mechanism of Death. In all nine cases, death was attributed to marked anoxemia resulting from a mechanical alveolar-capillary block. Major factors preventing adequate diffusion of oxygen include interstitial inflammation, pulmonary congestion, edema, and hyaline membrane formation. The septal inflammation, vascular congestion and edema increase the distance between the alveolar surface and the capillaries, thus decreasing the vital exchange of gases. The consequent hypoxia probably results in pulmonary hypertension putting a greater demand on the right ventricle. If cardiac insufficiency follows, hypoxia is intensified, initiating a lethal cycle. Mitral stenosis with its attendant alterations of pulmonary structure and function would tend to aggravate this already impaired diffusion system to a critical end point, rendering the maintenance of life impossible.

The influenza virus reaches the airways and lungs via inhalation and from these sites it enters the blood to be disseminated throughout the other tissues. It has been demonstrated in respiratory studies of the influenza virus that prior to its intracellular multiplication marked suppression of oxygen consumption in the host cells occurs [9]. This *in vivo* toxic factor may be partially responsible for the early hypoxia and cyanosis of these patients.

The presence of the hyaline membrane, with or without any of the other factors, offers a major barrier to the proper exchange of gases. Among the biophysical properties of this membrane, density is perhaps the most important one interfering with vital gaseous exchange. The membrane has been well labelled "asphyxial."

All these features, interstitial inflammation, edema and hyaline membrane formation, probably contribute to the stiffness and inelasticity of the lung producing a non-compliant and inefficient lung. The combination of these pulmonary factors and the viremia may account for the distinct lavender facial cyanosis, the rapid clinical deterioration of these patients and sudden death.

Morphologic Sequels of Influenzal Pneumonia. The potential or actual residua of an attack of influenzal pneumonia, although one of the facets of postinflammatory states, can only be interpreted from a speculative viewpoint.

With the diffuseness of the pulmonary changes a characteristic feature of these fatal instances of Asian influenza, the necrotizing septal changes as seen in Cases 3 and 6 if occurring in surviving instances of this pneumonia would probably leave only focal linear scars, difficult to label in terms of the original insult. An assumption can be made that if this fibrotic response is severe enough, it would be followed by distortion of the lung architecture with chronic impairment of pulmonary functions. But, long term follow-ups of patients afflicted with influenza in 1918 did not show an increase in incidence or severity of emphysema in the years immediately following that epidemic.

The bronchiolitis, probably both viral and bacterial in its inception and progress, would lead to some obliterative scars and might contribute to the genesis of focal emphysema. The structural change observed in the fatal cases of Asian influenza pneumonia, loosely labelled acute emphysema, is in reality an overdistention of the alveolar ducts and, unlike genuine emphysema, probably reversible.

SUMMARY

1. Nine patients with fulminant cases of Asian influenzal pneumonia have been examined by pathologic and virologic studies. The diagnosis in six of these nine cases was proved by isolation of the Asian strain of the influenza virus from the lungs, the remainder by the distinctive histologic findings.

2. Bacterial superinfection and its relation-

ship to viral infection is considered, in the light of suppression of microbial infections by antibiotic therapy and consequent clarification of the effects of the influenza virus upon lung tissue. The organisms recovered were Staphylococcus aureus, anaerobic streptococci and diphtheroids.

3. Antecedent conditions and contributory pathologic features were noted in the majority of these cases; their role as contributing to sudden death is discussed.

4. Anoxemia was thought to be the essential cause of death. The anoxemia was due to a mechanical alveolar-capillary block. Biophysical analysis by ultra soft x-rays indicates the density of the hyaline membrane, offering a mechanical block to gaseous exchange. Contributory causes were bacterial superinfection and the generalized toxemia.

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Studies on the Origin of Drug-Resistant Staphylococci in a Mental Hospital*

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IN studies in hospital wards which were heavily populated with coagulase-positive staphylococci resistant to several antibiotics, we have observed that treatment with tetracyclines and penicillin in new patients with susceptible staphylococci rapidly causes them to lose these organisms and acquire resistant strains from the environment, while similar patients who are untreated undergo this change very slowly. It was further found that although antibiotics influence the kind of staphylococci which are carried, the capacity to be a carrier appears to be independent of antimicrobial drugs [*1–3*].

In this report the preceding studies were extended by examining the mode of origin of reservoirs of resistant staphylococci by means of therapy experiments in a hospital ward where staphylococci were largely susceptible. The studies were particularly concerned with the effect of penicillin, tetracycline and erythromycin on the susceptibility of hospital staphylococci to drugs, on phage patterns, and on the capacity of patients to be staphylococcal carriers. The staphylococci described in this report were all obtained from nasal carriers and were characterized by production of coagulase and varying degrees of yellow pigmentation.

BACKGROUND INFORMATION

To illustrate the relatively high susceptibility to antibiotics of staphylococci carried by the mental patients and to show evidence, based on the distribution of phage patterns, that these patients had had little exposure to antibiotics, certain characteristics of 1,543 strains from this ward [*4*] are described in Figure 1 and Table I. For comparison similar data are presented from recent collections of staphylococci from Bellevue Hospital in New York [*1*] and the Veterans Administration Hospital in Nashville [*3*], and from a collection made at the Postgraduate Hospital in New York in a period largely before antibiotics were available [*1*]. The collections from the Bellevue and the Veterans Administration Hospitals are representative of a large number of hospitals in this country where antibiotics have been used heavily for a number of years. The Postgraduate Hospital collection was known to have had only a slight exposure to penicillin and to no other antibiotics.

It has been found that the use of antibiotics in hospitals is soon followed by an increase in staphylococci resistant to them. When more than one drug is used there is a great tendency for strains already resistant to one drug to become resistant to another. Furthermore, these strains are likely to be lysed by group III phages. In Figure 1 it may be seen that the cultures from the Bellevue and the Veterans Administration Hospitals showed a high percentage of resistance to penicillin, tetracycline, and lysis by group III phages, and examination of individual strains revealed that virtually every strain resistant to tetracycline (the most recently introduced drug) was also resistant to penicillin and classifiable into phage group III. Strains with these characteristics have been described as "hospital" staphylococci because of their occurrence in

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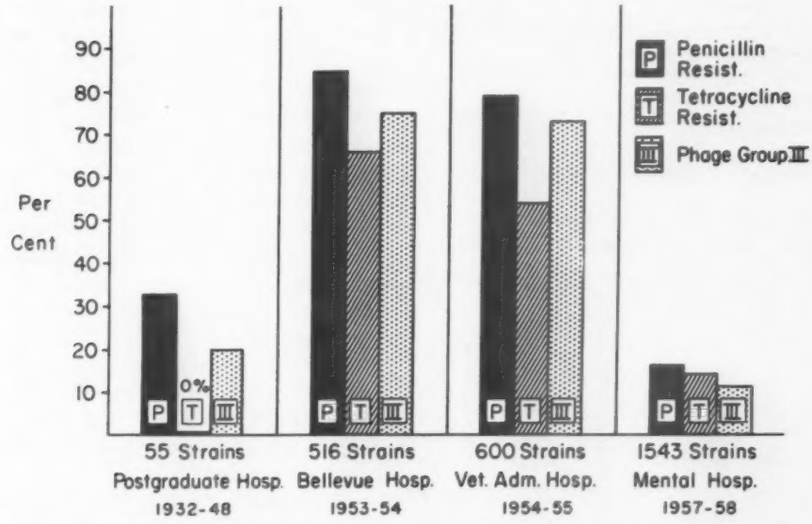

FIG. 1. Staphylococci from the mental hospital (from Knight et al. [4]), and those from Postgraduate Hospital collected largely before antibiotics were available, were quite susceptible to penicillin and tetracycline and were infrequently lysed by group III phages [1]. At Bellevue Hospital [1] and the Veterans Administration Hospital [2] they were resistant to these drugs and were predominantly lysed by group III phages.

these environments and their relative infrequency elsewhere. Although not illustrated, it was found that almost every strain resistant to the two antibiotics and lysed by group III phages was also resistant to streptomycin, a drug frequently used in these institutions. Erythromycin was infrequently used in the two hospitals at the time these collections were made and almost no strains resistant to it were found.

In Figure 1 it will be seen that the mental hospital and Postgraduate Hospital collections had few strains resistant to penicillin and tetracycline, and lysed by group III phages; moreover, it was known that more than half of the 30 per cent of strains at Postgraduate Hospital which were resistant to penicillin had been isolated in 1946 or later when this drug had been used. Since it was known that the tetracyclines and penicillin were occasionally used at the mental hospital it could be safely estimated that the small percentage of resistant strains there resulted, at least in part, from drug exposure. As at the Bellevue and the Veterans Administration Hospitals, erythromycin had hardly been used at the mental hospital and no strains in this collection were resistant to it.

TABLE I

FREQUENCY OF PENICILLIN-RESISTANT, TETRACYCLINE-RESISTANT AND PHAGE GROUP III STAPHYLOCOCCI IN COLLECTIONS FROM FOUR DIFFERENT HOSPITALS

Hospital	Penicillin-Resistant (10 mcg./ml. or greater)	Tetracycline(s)-Resistant (25 mcg./ml. or greater)	Phage Group III (%)
Postgraduate Hospital, New York, 1932–1948, 55 strains	33%	0%	20%
Bellevue Hospital, New York, 1953–1954, 516 strains	85%	66%	75%
Veterans Administration Hospital, Nashville, 1954–1955, 600 strains	79%	54%	73%
Mental Hospital, Nashville, 1958, 1,543 strains	16%	14%	11%

EXPERIMENTAL SUBJECTS AND METHODS OF STUDY

Mental Hospital Patients. The subjects in the study were all women with serious chronic mental illness who had resided on the ward of a mental hospital for many years. Most were in the age range of thirty-five to fifty years. They were ambulatory but were allowed to leave the ward for only short periods during the day. More than 90 per cent regularly received treatment with a tranquilizing drug, usually chlorpromazine. Review of their hospital records for the preceding two years revealed only occasional use of common antimicrobial drugs. No cases of serious staphylococcal infection had been recorded in this period and none occurred during the course of the study.

TABLE II
EFFECT OF ERYTHROMYCIN ON NASAL CARRIER RATES OF COAGULASE-POSITIVE STAPHYLOCOCCI

Date	Treated Carriers (6)		Untreated Carriers (5)	
	No. of Cultures	Per cent Positive	No. of Cultures	Per cent Positive
1957:				
11/15	6	83.3	5	60.0
11/18	6	100.0	5	100.0
11/19	6	83.3	5	60.0
11/20	6	83.3	5	60.0
11/21	5	100.0	5	100.0
11/25	6	66.6	5	60.0
11/26	6	66.6	5	40.0
11/27	6	83.3	5	100.0
11/28	6	66.6	5	80.0
12/2	6	16.6	5	100.0
12/3	6	16.6	5	80.0
12/4	6	0.0	5	60.0
12/5	6	0.0	5	100.0
12/7	5	20.0	4	75.0
12/9	6	0.0	5	100.0
12/10	6	33.3	5	100.0
12/11	6	33.3	5	100.0
12/12	6	33.3	5	100.0
12/13	6	33.3	5	80.0
12/16	6	33.3	5	100.0
12/17	6	33.3	5	100.0
12/19	6	83.3	5	100.0
12/20	6	66.6	5	80.0
12/23	5	80.0	5	80.0
12/24	5	60.0	5	80.0
12/26	5	100.0	5	80.0
12/27	6	100.0	5	100.0
12/30	6	100.0	5	80.0
12/31	6	83.3	5	80.0
1958:				
1/2	6	66.6	5	80.0
1/3	6	83.3	4	75.0
1/6	6	100.0	5	60.0
1/7	6	100.0	5	100.0
1/8	6	83.3	5	80.0
1/9	6	100.0	5	100.0
1/10	6	83.3	5	100.0

NOTE: Treatment consisted of erythromycin 1.0 gm. daily from November 27 to December 6, 1957.

The ward had an average census of sixty-two patients of whom about fifty permitted the regular nasal swabbings required in the study. No more than five or six patients were admitted or discharged from the ward annually. Preliminary nasal swabbings indicated that most patients could be divided into groups of carriers—those with regularly positive cultures, and non-carriers, those with none or only sporadic positive cultures—and that only a small percentage of patients changed status in any one month [*4*]. The size of the treatment and non-treatment groups among carriers and non-carriers was arbitrarily decided upon, but the assignment of patients to them was determined by use of a table of random numbers. It was elected to treat the same carriers who received penicillin with tetracycline so that no randomization was needed for this experiment. As a consequence of suppression of the carrier state in a few patients by penicillin, fewer carriers were available to be treated with tetracycline.

Nasal Swabbing. Sterilized, commercially prepared cotton applicators 5 inches in length were moistened in sterile broth and used immediately to swab both anterior nares by a circular motion. The swabs were placed in Wassermann tubes containing 3 ml. of sterile broth and were taken to the laboratory without delay.

Enumeration of Staphylococcal Colonies from Swabs. Inoculated swabs in tubes were shaken for five minutes in a Kahn shaker, the swabs discarded, and serial tenfold dilutions made of the remaining broth. Using an 0.2 ml. pipette, exactly 0.1 ml. each of undiluted 1:10, and 1:100 dilutions of the inoculum were dropped on the surface of an agar medium containing tryptic digest of casein and soya* which had been dried for forty-eight hours at 37°c. The plates were incubated overnight at 37°c. and then held for an additional day at room temperature to allow development of pigment. Pigmented colonies, of which a representative produced coagulase, were counted and a subculture was preserved for the study. By this method approximately 55 to 60 per cent of patients were found to be nasal carriers of pigmented, coagulase-positive staphylococci in any one culture period when treatment was not being given [*4*].

The number of colonies per plate multiplied by the appropriate dilution factors gave the number of colonies per swab. The lowest possible positive value was thirty per swab but most carriers had several thousand colonies per swab. An analysis of this procedure will be described elsewhere [*5*], and in this report quantitative swab counts are shown only for comparison with conventional carrier rate determinations made from the same swabs.

Coagulase Test. A loopful of the colony to be tested was mixed with 0.5 ml. of sterile blood bank plasma and incubated in a water bath at 37°c. for three hours. Any recognizable clot formation in this period was considered evidence of coagulase production.

Agar Dilution Test for Antimicrobial Susceptibility. Broth inoculums were added to agar plates containing specified concentrations of antimicrobial drugs by means of a capillary tube, and growth was read after overnight incubation. Since one or two colonies occasionally grew in high concentrations of drug, which on subsequent tests showed no growth, this slight

* Trypticase soy agar, Baltimore Biological Laboratories, Baltimore, Maryland.

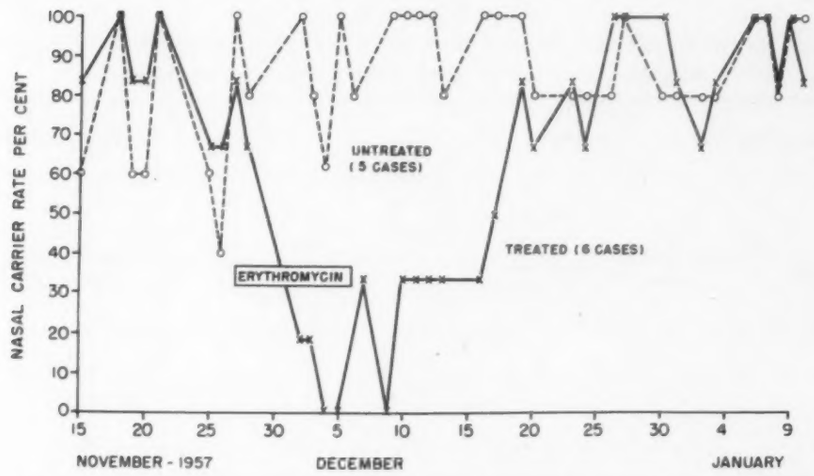

FIG. 2. Effect of erythromycin on nasal staphylococcal carriers. Erythromycin completely suppressed the carrier state while untreated carriers did not change. After treatment the carrier rate returned to pretreatment values in about two weeks.

growth was disregarded in reading the plates. Cultures inhibited by 1.0 μg. per ml. of penicillin, 1.6 of erythromycin, and 6.2 of tetracycline were considered susceptible. Those not inhibited by these concentrations were considered to be resistant.

Phage Typing. Stock phages were propagated in heart infusion broth containing 0.004 moles per L. of added calcium [6]. Staphylococci to be typed were exposed on plates to suspensions of the phages in the tenfold dilution of the stock which caused complete lysis of the propagating strain. If no lysis of unknown strains occurred they were re-exposed to a 1,000-fold concentration of the first testing dilution except for the few phages in which the titer of stocks was not sufficient to allow this concentration. In these cases undiluted stock was used.

The degree of lysis was read on a scale of 1 to 4 plus. One plus indicated at least twenty discrete plaques per spot; 2 plus, fifty or more plaques; 3 plus, semiconfluent lysis; and 4 plus, confluent lysis. A large majority of the greater than 90 per cent of strains which were lysed by one of the two test dilutions gave 3- or 4-plus lysis. About 50 per cent were lysed by the first test dilution and slightly over 40 per cent by the second. Staphylococci were classified into the following categories in the present investigation: 52–80; other group I cultures; phage group II; phage group III; those that did not react with either testing dilution (no type or untypable); and those of mixed patterns (miscellaneous). The cultures which reacted with group I phages 52–80 also regularly reacted with less intensity with phages 79, 81, 47c and 42e.

The following phages* were used in this investigation: group I, 29, 52, 52a, 79, 80; group II, 3a, 3b, 3c, 55; group III, 6, 7, 42e, 47, 53, 54, 70, 73, 75, 77, 42b, VA_4, 42d, 47c; and Unclassified, 81, 44a.

* The phages and their propagating strains were obtained from Dr. John Blair, Hospital for Joint Diseases, New York City.

RESULTS

Nasal swabbings were made regularly of patients in the ward beginning in October 1957, and were continued through August, 1958. In this setting selected patients were given courses of treatment with erythromycin, penicillin and tetracycline in order to judge the effect of these agents on various aspects of the staphylococcal carrier state.

Erythromycin Experiment. In Figure 2 and Table II are shown the carrier rates of patients who were treated with erythromycin. It may be seen that after four days of treatment staphylococci had disappeared in all of the treated carriers. They remained essentially negative for staphylococci during the remainder of treatment and for about nine days thereafter. At this time the carrier rate increased rapidly to pretreatment values. The untreated carriers continued to carry staphylococci with the same frequency throughout this period. Not shown in Figure 2 are two groups of non-carriers, one of which received erythromycin, whose status remained unchanged during this experiment. During the week following treatment five cultures from two treated carriers and one from an untreated carrier were found to be resistant to erythromycin. Three of these were phage type 52–80 and three were untypable strains. Cultures made subsequently were all susceptible to this agent.

TABLE III

CARRIER RATES OF COAGULASE-POSITIVE STAPHYLOCOCCI FROM NASAL SWABS OF MENTAL PATIENTS BEFORE AND AFTER TREATMENT WITH PENICILLIN

Date (1958)	Carriers				Non-Carriers		
	No. Cultured	All Staphylococci (per cent)	Penicillin-Resistant Staphylococci (per cent)	Mean of Logs of Swab Counts	No. Cultured	All Staphylococci (per cent)	Mean of Logs of Swab Counts
3/10	16	100.0	25.0	4.06	15	26.6	.38
3/12	16	93.7	6.2	3.74	15	33.3	.54
3/14	16	88.0	6.2	3.23	15	20.0	.36
3/17	16	100.0	12.5	3.91	15	20.0	.25
3/19	16	93.7	12.5	3.47	14	28.5	.22
3/21	14	100.0	14.2	3.11	15	6.6	.17
3/24	16	100.0	25.0	4.06	15	6.6	.39
3/26	15	93.3	0.0	3.33	15	0.0	0
3/28	16	100.0	12.5	4.07	15	0.0	0
3/31	16	100.0	0.0	4.12	15	0.0	0
4/2	16	81.2	12.5	3.44	15	6.6	.17
4/3	16	68.7	12.5	2.55	15	13.3	.45
4/4	16	75.0	6.2	2.33	15	6.6	.29
4/7	16	61.8	18.7	2.15	15	6.6	.27
4/8	16	61.8	31.2	1.88	15	0.0	0
4/9	16	61.8	18.7	1.99	15	6.6	.18
4/10	16	75.0	37.5	2.11	15	20.0	0
4/11	16	75.0	68.7	2.44	15	13.3	.30
4/14	15	53.3	53.3	2.03	14	7.1	.34
4/15	16	75.0	37.5	1.98	15	6.6	.34
4/16	16	50.0	50.0	1.73	15	6.6	.14
4/17	16	37.5	37.5	1.55	15	0.0	0
4/18	16	50.0	37.5	2.08	15	6.6	.24
4/19	15	40.0	33.3	1.48	15	0.0	0
4/21	16	68.7	56.3	2.33	15	6.6	.37
4/22	16	50.0	50.0	1.80	15	6.6	.46
4/23	16	75.0	50.0	1.56	15	13.3	.34
4/24	16	50.0	50.0	1.88	15	6.6	.44
4/25	16	75.0	37.5	1.86	15	0.0	0
4/26	16	62.5	50.0	1.87	15	6.6	.52
4/28	15	60.0	33.3	1.93	15	6.6	.45
4/29	16	62.5	62.5	2.18	15	6.6	.68
4/30	16	75.0	56.3	1.93	15	0.0	0
5/1	16	75.0	56.3	2.03	15	6.6	.21
5/2	16	37.5	31.2	1.52	15	0.0	0
5/3	16	62.5	45.0	2.19	15	6.6	.32
5/5	16	56.6	45.0	2.41	15	13.3	.54
5/6	15	75.0	53.3	2.65	15	6.6	.38
5/7	15	53.3	53.3	1.87	15	0.0	0
5/8	15	60.0	53.3	2.44	15	6.6	.36
5/9	15	60.0	46.6	2.21	15	0.0	0
5/10	15	53.3	53.3	2.02	15	6.6	.22
5/12	15	60.0	53.3	2.64	15	6.6	.39
5/13	15	60.0	....	2.56	15	0.0	0
5/14	16	56.2	....	2.26	15	6.6	.18
5/15	16	56.2	....	2.36	14	6.6	0
5/16	16	56.2	56.6	2.42	15	6.6	.19
5/19	16	62.5	56.6	2.33	15	20.0	.51
5/20	16	56.2	31.2	2.33	15	13.3	.23
5/21	16	62.5	45.0	2.47	15	6.6	.18

TABLE III (*Continued*)

CARRIER RATES OF COAGULASE-POSITIVE STAPHYLOCOCCI FROM NASAL SWABS OF MENTAL PATIENTS BEFORE AND AFTER TREATMENT WITH PENICILLIN

Date (1958)	Carriers				Non-Carriers		
	No. Cultured	All Staphylococci (per cent)	Penicillin-Resistant Staphylococci (per cent)	Mean of Logs of Swab Counts	No. Cultured	All Staphylococci (per cent)	Mean of Logs of Swab Counts
5/23	16	62.5	45.0	2.49	15	6.6	.31
5/26	16	68.7	50.0	2.88	15	0.0	0
5/27	16	68.7	61.8	2.89	14	6.6	.33
5/28	16	68.7	61.8	2.77	15	13.3	.32
5/29	16	68.7	50.0	2.79	15	13.3	.25
5/31	15	66.6	56.6	2.43	15	0.0	0
6/2	16	62.5	50.0	2.28	15	13.3	.42
6/3	16	62.5	61.8	2.81	15	0.0	0
6/4	16	75.0	61.8	2.91	15	6.6	.31
6/5	16	75.0	61.8	3.07	15	6.6	.34
6/6	16	75.0	56.6	2.81	14	6.6	.28
6/9	16	75.0	56.6	3.08	15	0.0	0
6/11	16	68.7	56.6	3.14	15	6.6	.29
6/13	16	68.7	37.5	2.53	15	6.6	.25
6/16	16	56.2	50.0	2.29	15	13.3	.43
6/18	16	62.5	18.7	2.54	15	6.6	.21
6/20	16	75.0	31.2	3.18	15	13.3	.41
6/23	15	66.6	50.0	2.63	15	13.3	.40

NOTE: Treatment consisted of penicillin, 1.5 million units daily, orally, from April 1 to May 13.

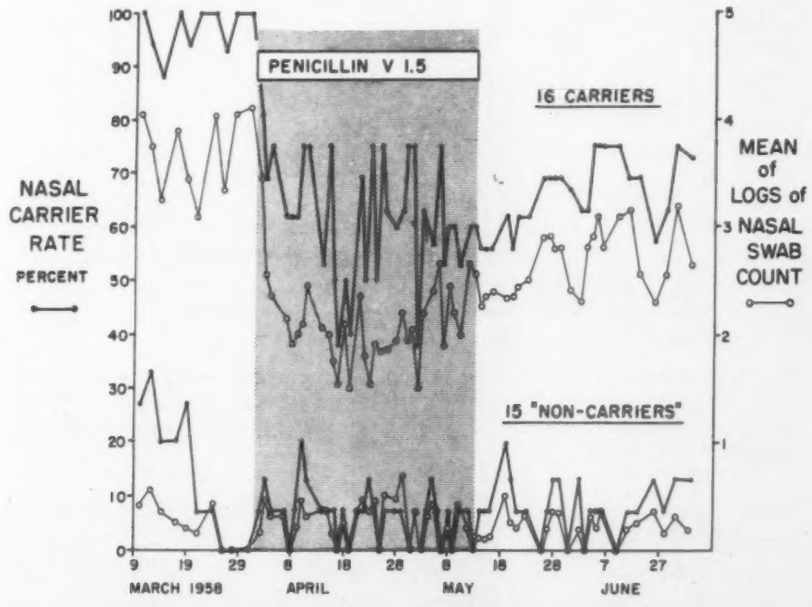

FIG. 3. Treatment with penicillin reduced carrier rates to about 60 per cent from 100 per cent. Nasal swab counts (see text) dropped to 10^2 from values ranging from 10^3 to 10^4. Treated non-carriers were not influenced. Carrier rates and counts slowly rose after treatment.

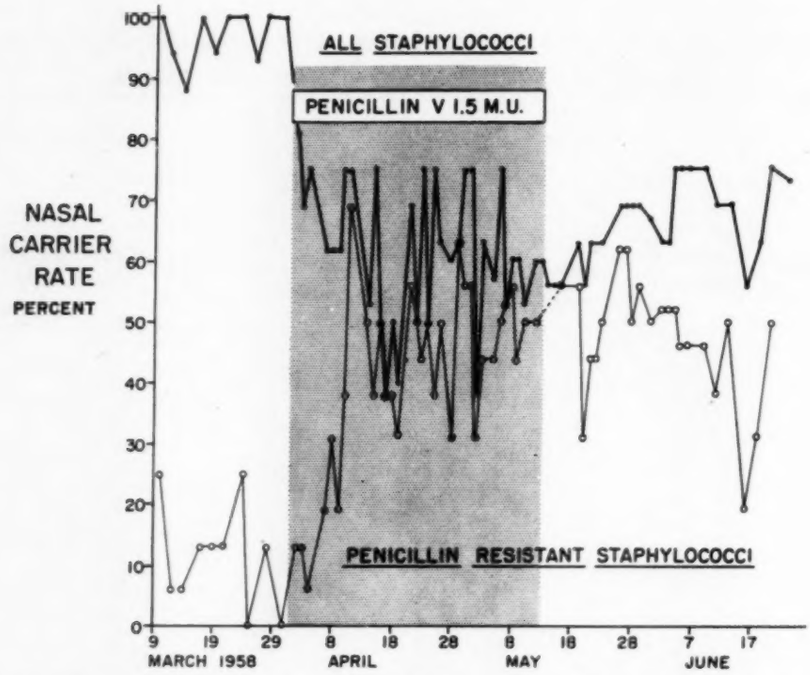

FIG. 4. The fall of carrier rates to about 60 per cent with penicillin was found to be limited at this point by an increase in strains resistant to penicillin. After treatment these strains gradually disappeared.

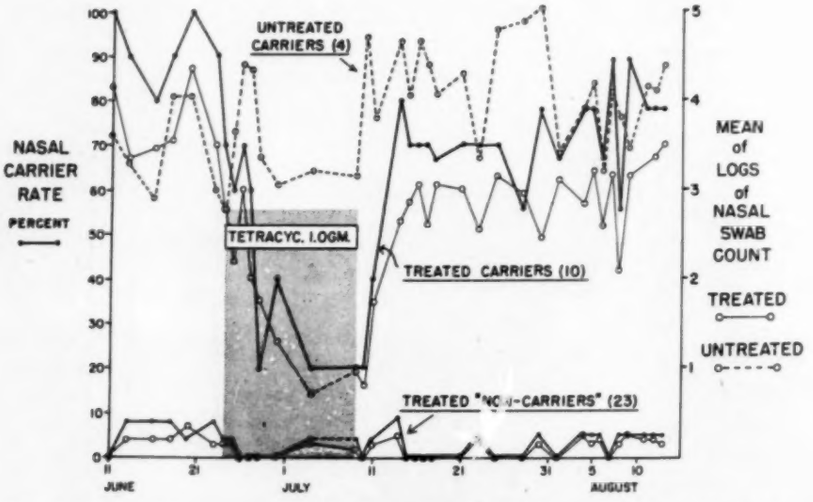

FIG. 5. Tetracycline caused a reduction in the carrier rate of treated carriers from 100 to 20 per cent. The rate returned almost to pretreatment values after treatment. Nasal swab counts paralleled carrier rates. Treated non-carriers were not appreciably influenced.

Penicillin Experiment. Five months after completing treatment with erythromycin, sixteen carriers and fifteen non-carriers were treated with oral penicillin V for six weeks. In Figure 3 and Table III are shown the nasal carrier rates and the quantitative nasal swab counts from these patients. It may be seen that the carrier rate and the mean swab count in the treated carriers promptly dropped to about half of their pretreatment values. Following this there was a period of several days of wide fluctuation in both the swab count and the carrier rate. Near the end of treatment, however, they stabilized and began a slow increase which was con-

TABLE IV

CARRIER RATES OF COAGULASE-POSITIVE STAPHYLOCOCCI FROM NASAL SWABS OF MENTAL PATIENTS BEFORE, DURING AND AFTER TREATMENT WITH TETRACYCLINE

	Treated Carriers					Non-Treated Carriers				Treated Non-Carriers		
Date (1958)	No. Cultured	All Staphylococci (per cent)	Tetracycline Resistant Staphylococci (per cent)	Tetracycline and Penicillin Resistant Staphylococci (per cent)	Mean of Logs of Quantitative Counts—All Staphylococci	No. Cultured	All Staphylococci (per cent)	Tetracycline Resistant Staphylococci (per cent)	Mean of Logs of Quantitative Counts	No. Cultured	All Staphylococci (per cent)	Mean of Logs of Quantitative Counts—All Staphylococci
6/11	10	100.0	0.0	0.0	4.14	4	100.0	0.0	3.61	23	0.0	0
6/13	10	90.0	0.0	0.0	3.36	4	100.0	0.0	3.31	23	8.6	.18
6/16	10	80.0	0.0	0.0	3.43	4	75.0	0.0	2.92	23	8.6	.22
6/18	10	90.0	0.0	0.0	3.56	4	100.0	0.0	4.05	23	8.6	.18
6/20	10	100.0	0.0	0.0	4.37	4	100.0	0.0	4.04	23	4.3	.34
6/23	10	90.0	0.0	0.0	3.49	4	100.0	0.0	3.02	23	8.6	.16
6/24	10	70.0	10.0	0.0	2.81	4	50.0	0.0	2.82	23	4.3	.14
6/25	10	60.0	0.0	0.0	2.20	4	50.0	0.0	3.64	23	4.3	.13
6/26	10	70.0	....	....	2.99	4	100.0	....	4.41	23	0.0	0
6/27	10	60.0	10.0	10.0	2.02	4	100.0	0.0	4.33	22	0.0	0
6/28	10	20.0	0.0	0.0	1.73	4	75.0	0.0	3.33	23	0.0	0
6/30	10	40.0	10.0	0.0	1.27	4	75.0	0.0	3.02	23	0.0	0
7/3	10	20.0	20.0	20.0	.69	4	75.0	0.0	3.18	23	4.3	.16
7/9	10	20.0	20.0	20.0	.93	4	75.0	0.0	3.14	23	4.3	.10
7/10	10	20.0	20.0	20.0	.82	4	100.0	0.0	4.70	23	0.0	0
7/11	10	40.0	20.0	20.0	1.74	4	75.0	0.0	3.81	23	4.3	.17
7/14	10	80.0	60.0	50.0	2.67	4	100.0	25.0	4.66	23	8.6	.25
7/15	10	70.0	60.0	60.0	2.83	4	100.0	0.0	4.04	23	0.0	0
7/16	10	70.0	40.0	40.0	3.05	4	100.0	25.0	4.65	23	0.0	0
7/17	10	70.0	50.0	50.0	2.62	4	100.0	25.0	4.41	23	0.0	0
7/18	9	66.6	55.5	56.0	3.04	4	100.0	25.0	4.07	23	0.0	0
7/21	10	70.0	60.0	50.0	2.98	4	100.0	0.0	4.31	23	0.0	0
7/23	10	70.0	30.0	30.0	2.53	4	50.0	0.0	3.33	22	4.5	.19
7/25	10	70.0	50.0	40.0	3.15	4	100.0	25.0	4.81	22	0.0	0
7/28	9	55.5	22.2	22.0	2.93	4	100.0	0.0	4.92	22	0.0	0
7/30	9	77.7	33.3	33.0	2.45	4	100.0	0.0	5.23	22	4.5	.16
8/1	9	66.6	22.2	22.0	3.12	4	75.0	0.0	3.42	21	0.0	0
8/4	9	77.7	44.4	33.0	2.85	4	100.0	25.0	3.89	22	4.5	.24
8/5	9	77.7	66.6	66.0	3.21	4	100.0	0.0	4.19	22	4.5	.14
8/6	9	66.6	33.3	33.0	2.57	4	75.0	....	3.19	22	4.5	.18
8/7	9	88.8	66.6	44.0	3.13	4	100.0	....	4.09	22	0.0	0
8/8	9	55.5	33.3	22.0	2.11	4	75.0	....	3.81	22	4.5	.14
8/9	9	88.8	22.2	11.0	3.14	4	75.0	....	3.43	22	4.5	.25
8/11	9	77.7	44.4	44.0	3.35	4	50.0	....	4.15	22	4.5	.18
8/12	9	77.7	22.2	12.0	3.49	4	100.0	....	4.12	22	4.5	.19
8/13	9	77.7	....	....	....	4	100.0	....	4.39	22	4.5	.17

NOTE: Tetracycline 1.0 gm. daily, orally, June 24 to July 8 inclusive.

tinued during several weeks of observation after treatment. The carrier rate and mean swab count of non-carriers showed no evidence of effect during this experiment.

An explanation was sought for the failure of the carrier rate and quantitative count in treated carriers to reduce with penicillin as they had with erythromycin. The reason for this became clearly evident when the resistance of the cultures to penicillin was determined. This is described in Figure 4 and Table III. By the ninth day of treatment the carrier rate of strains resistant to this agent had increased from approximately 10 per cent to 50 per cent, so that these strains accounted for virtually all of the positive cultures during most of the period of treatment with penicillin and for about three weeks thereafter. At this time the frequency of strains resistant to penicillin diminished while carrier rates for all staphylococci ascended, thus establishing a significant trend toward the pretreatment situation.

Tetracycline Experiment. Forty days after stopping penicillin ten of the sixteen carriers treated with penicillin who were still carrying staphylococci were given two weeks treatment with tetracycline. In addition, twenty-three non-carriers were given the drug. Four carriers were observed without treatment. These data are described in Figure 5 and Table IV. The carrier rate of the treated carriers dropped within a few days to about 20 per cent and the mean swab

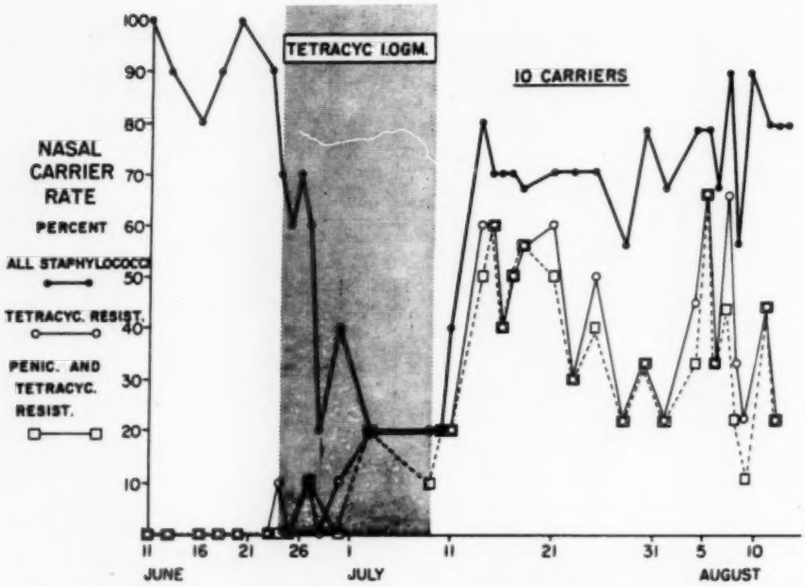

FIG. 6. The fall of the carrier rate with tetracycline was found to be halted at 20 per cent by increase in the carrier rate of resistant strains to this value (clear circles on graph). It was further found that resistance to tetracycline had developed almost exclusively in strains resistant to penicillin, so that "double resistant" strains appeared for the first time.

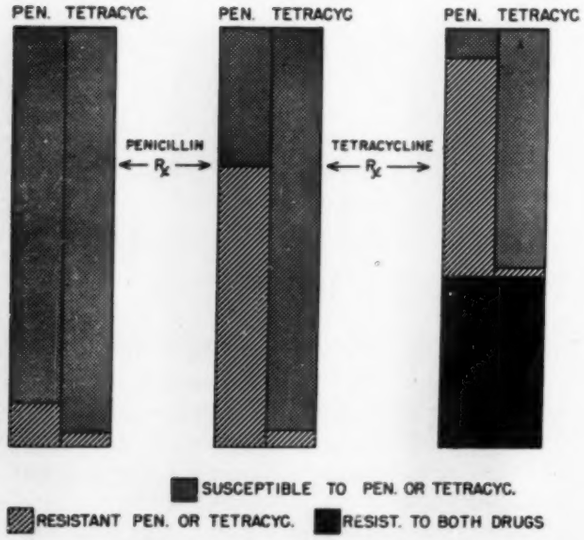

FIG. 7. Collections of staphylococci made before penicillin, after it and before tetracycline, and after tetracycline increased their resistance to penicillin from 10 to 67 to 93 per cent. Resistance to tetracycline increased from 4 to 43 per cent after its use. Most strains resistant to tetracycline were also resistant to penicillin. This suggested that strains resistant to penicillin possessed greater capacity to become resistant to tetracycline—and because of this linkage of penicillin and tetracycline resistance, tetracycline caused penicillin resistance to increase.

count diminished to ten colonies per swab. Immediately after treatment was stopped both the carrier rate and the mean swab count increased rapidly almost to pretreatment values. It was evident that this response was intermediate between the effect of erythromycin and penicillin. Untreated carriers and untreated non-carriers showed no definite changes in this period.

Observation of drug resistance during this period revealed, as with penicillin, that failure to suppress the carrier state completely resulted from the appearance of resistant strains (Fig. 6, Table IV). Furthermore, it was found that development of resistance to tetracycline had been almost exclusively restricted to strains already resistant to penicillin. (Fig. 6.)

Drug Resistance and Phage Patterns of Staphylococci Before Penicillin, After Penicillin and Before Tetracycline, and After Tetracycline. For the purpose of estimating the effect of antibiotics on drug resistance and phage patterns, a study was made of collections of staphylococci isolated from all of the patients on the ward in approximately two-week periods in the intervals before and after treatment with penicillin and after tetracycline. The changes in susceptibility to penicillin and tetracycline are shown in Figure 7 and Table V. These data show that following

TABLE V

CHARACTERISTICS OF COAGULASE-POSITIVE STAPHYLOCOCCI FROM NASAL SWABS OF MENTAL PATIENTS BEFORE AND AFTER PENICILLIN AND AFTER PENICILLIN AND TETRACYCLINE

Antimicrobial Susceptibility	Phage Pattern	Before Penicillin		After Penicillin and Before Tetracycline		After Penicillin and Tetracycline	
		No. of Strains	Per cent	No. of Strains	Per cent	No. of Strains	Per cent
Penicillin-susceptible	52–80	95	54.6	11	11.1	0	0.0
	III	12	6.9	1	1.0	0	0.0
	II	27	16.1	12	12.0	5	7.0
	I	0	0.0	2	2.0	0	0.0
	No type	23	13.2	6	5.9	0	0.0
	Miscellaneous	0	0.0	1	1.0	0	0.0
	Subtotal	157	90.8	33	33.0	5	7.0
Penicillin-resistant	52–80	0	0.0	0	0.0	0	0.0
	III	3	1.7	19	17.8	19	26.4
	II	7	4.0	16	15.8	15	20.8
	I	1	0.6	2	1.9	0	0.0
	No type	4	2.3	28	27.5	33	45.8
	Miscellaneous	1	0.6	4	4.0	0	0.0
	Subtotal	16	9.2	69	67.0	67	93.0
	Total	174	100.0	102	100.0	72	100.0
Tetracycline-susceptible	52–80	92	52.9	11	10.8	0	0.0
	III	15	6.0	19	18.6	13	18.0
	II	35	20.1	28	27.4	13	18.0
	I	1	0.6	3	3.0	0	0.0
	No type	26	14.9	31	30.4	15	21.0
	Miscellaneous	1	0.6	6	5.8	0	0.0
	Subtotal	170	97.7	98	96.0	41	57.0
Tetracycline-resistant	52–80	3	1.7	1	1.0	0	0.0
	III	0	0.0	0	0.0	6	8.3
	II	0	0.0	0	0.0	7	9.7
	I	0	0.0	0	0.0	0	0.0
	No type	1	0.6	3	3.0	18	25.0
	Miscellaneous	0	0.0	0	0.0	0	0.0
	Subtotal	4	2.3	4	4.0	31*	43.0
	Total	174	100.0	102	100.0	72	100.0

NOTE: Treatment consisted of penicillin V to sixteen carriers, 1.5 million units daily, orally, 4/1 to 5/13/58; tetracycline, 1.0 gm. daily, 6/25 to 7/8/58. Collection period of cultures: before penicillin, 3/17 to 3/31; after penicillin, 6/11 to 6/23; after penicillin and tetracycline, 7/25 to 8/5/58.

* Thirty of these cultures were also resistant to penicillin.

treatment with penicillin there was a pronounced increase in the percentage of strains resistant to this drug, confirming the observation of this event in Figure 4. Resistance to tetracycline was not increased by treatment with penicillin. Following treatment with tetracycline, resistance to this agent increased from 4 to 43 per cent—again confirming previous observations. In addition, it was found that resistance to penicillin, which reached 67 per cent after use of this drug, further increased to 93 per cent after treatment with tetracycline. Moreover, for the first time in the nine months in which regular cultures had been made on the

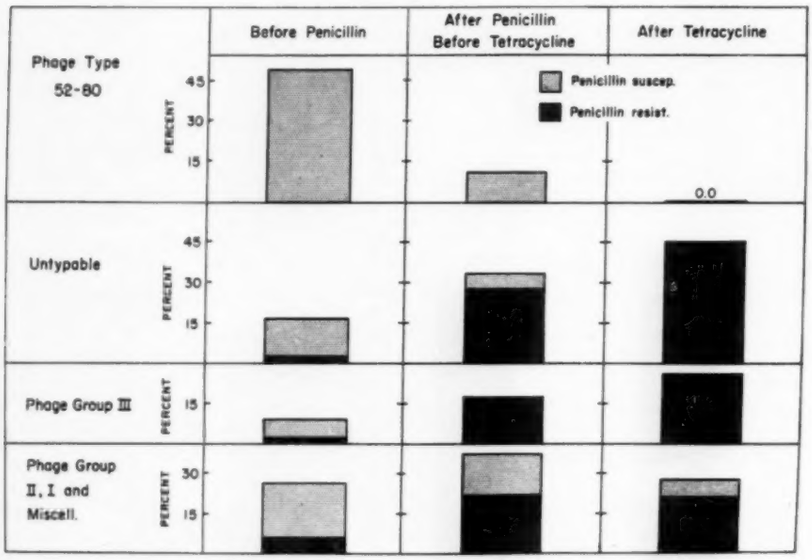

FIG. 8. Penicillin and tetracycline caused the complete disappearance of the more than 50 per cent of phage group I, 52–80, strains originally present in the ward. Reciprocally, untypable and phage group III strains resistant to penicillin increased from less than 5 per cent to constitute about three-fourths of the cultures obtained after tetracycline. The strains resistant to penicillin and tetracycline (Fig. 7) were about a half untypable and a fourth each phage group III and phage group II.

ward, strains resistant both to tetracycline and penicillin appeared. (Fig. 7.) They constituted 41.6 per cent of all the strains in the third collection and thirty of the thirty-one strains resistant to tetracycline.

Analysis of the distribution of phage patterns in the three collections showed three important shifts which clearly suggested a drug effect. (Fig. 8.) Strains of the group I phage type, 52–80, susceptible to penicillin and tetracycline, which constituted more than half of the strains collected before penicillin, were reduced to about 10 per cent after treatment with this drug, and completely disappeared following tetracycline. None of this large number of strains became resistant to penicillin and the only strain apparently resistant to tetracycline was isolated before it had been used. (Table V.) The failure of these strains to develop resistance to penicillin and tetracycline was of particular interest since they showed minor reactions with phages 79, 81, 47c and 42b, indicating a significant relationship to resistant strains of similar patterns, which have been responsible for a large number of outbreaks of staphylococcal infection in hospitals [7].

In a reciprocal manner strains designated "untypable," which indicates failure to lyse when exposed to a concentrated suspension of each of the twenty-five typing phages, increased progressively in the three collections from 15.6 to 33.4 to 45.8 per cent. Probably more significantly, untypable strains resistant to penicillin constituted only 2.3 per cent of the collection before treatment with penicillin; after penicillin they increased to 27.5 per cent and following tetracycline they reached 45.8 per cent, and at this time no untypable strains susceptible to penicillin remained. Finally, it was noted that there was a similar but lesser increase in the proportion of strains of phage group III patterns resistant to penicillin in the three successive collections. Strains of phage group I (other than 52–80), II and those of the miscellaneous category showed increasing resistance, but quantitatively their participation was less than that of the other groups.

In Table V it may be seen that the percentage shifts to resistance to tetracycline after treatment with this drug were smaller, possibly as a result of the shorter period of use of this drug. Nevertheless, it may be seen that more than half of the 43 per cent of strains resistant to tetracycline were untypable and all but one were also resistant to penicillin.

COMMENTS

This investigation has revealed several aspects of the mechanism by which antimicrobial therapy can cause an increase in resistance of the staphylococcal flora of a hospital ward. It was observed that the carrier state was completely suppressed by treatment with erythromycin, and this was correlated with uniform susceptibility of staphylococci on the ward to the drug before treatment, and with failure of resistant strains to appear during treatment (although a few resistant strains were isolated later). In contrast, with penicillin the carrier rate dropped from 100 to about 60 per cent and the reduction was found to be halted at this point because of the increase in carrier rates of resistant strains from 10 per cent before treatment to about 50 per cent. Thus, resistant strains accounted for nearly all of the positive cultures during this period. There were fewer strains resistant to tetracycline before this drug was used—only about 4 per cent— and use of this drug caused a drop in the carrier rate to 20 per cent before it was limited by the appearance of resistant strains. Thus erythromycin was the most effective suppressive agent, tetracycline was intermediate, and penicillin was least effective.

A most interesting aspect of the increase in resistance of staphylococci in this study was the observation that strains resistant to penicillin were almost exclusively selected to become resistant to tetracycline. If tetracycline had acted in a random manner, strains susceptible to penicillin would have constituted about one-third of those resistant to tetracycline. As it was, only one strain resistant to tetracycline was susceptible to penicillin. The most logical explanation of this occurrence appears to be that strains which have become resistant to penicillin have a greater capacity to become resistant to antibiotics—possibly due to a greater mutative tendency, and the use of tetracycline led to the selection of tetracycline-resistant strains from this group. Moreover, the studies of the distribution of various phage groups at intervals during the treatment experiments revealed evidence which we believe can best be interpreted as indicating a greater capacity of strains of certain phage patterns to survive in the presence of drug. Thus, among the small number of strains resistant to penicillin before this drug was used only a small percentage were of "no type," or phage group III, yet after treatment these strains had gained a clear predominance among the resistant strains in the ward.

We have assumed that the small proportion of resistant strains of the phage patterns which were present before penicillin was used were the source of the larger numbers of these resistant strains after treatment. This assumption seems preferable to suggesting that susceptible strains of these types also present in the ward before treatment had become resistant. This view was strengthened by the observation that the strains of phage type 52–80 which were susceptible to penicillin and tetracycline (with one possible exception) disappeared from the ward following treatment with these agents. Presumably, with erythromycin, when no resistant strains were available resistance would develop in phage types most adaptable to drug exposure. In this study the few resistant strains which appeared were equally divided between "no type" and 52–80.

As previously described, cultures from patients on wards where antibiotics have been heavily used have characteristically shown a predominance of group III phage patterns and resistance to antibiotics. In the present study the predominant resistant group was "no type." We have considered that the relatively brief exposure of the present patients to antimicrobial drugs might not be sufficient to cause a predominance of group III strains, but the fact that it was sufficient to cause another group to be predominant is best interpreted as evidence of an unexplained inconsistency between the present study and the results after ordinary therapy. It is possible that further changes with continued drug exposure may take place and such experiments are now underway.

It has long been a question of importance whether or not antimicrobial treatment would cause patients to become carriers of staphylococci, either by some influence on the patient which would make his tissues more susceptible to superficial infection with staphylococci, or by endowing staphylococci made drug-resistant with a property of greater invasiveness. In previous studies we have described data which showed no increase in carrier rates of staphylococci in patients receiving tetracycline or penicillin [*2*]. In the present study further support for these findings was obtained when it was seen that a large number of non-carriers (patients with only occasional positive cultures) treated with penicillin and tetracycline did not have

more positive cultures during or after treatment than they had before treatment.

Since the treated carriers were nearly 100 per cent positive before treatment, an increase in these patients could not be demonstrated. It would have been possible for carriers to carry larger numbers of staphylococci but the quantitative swab counts did not suggest such an occurrence. Lepper and his associates have reported that treatment with chlortetracycline was associated with increase in carrier rates of staphylococci [*8*]. We have not examined this drug in this respect but the general question is of such importance that efforts should be made with all drugs to determine whether or not they can alter the equilibrium between staphylococci and man to cause the carrier state to develop.

The implications of these studies concerning control of drug resistance by withholding certain agents from use are not encouraging. Considering the frequent valid indications for the use of penicillin and tetracyclines, it is difficult to imagine a program of restricted use of these agents which would result in less use of the drugs than occurred in the mental hospital. And despite this low rate of usage strains resistant to penicillin and tetracycline appeared in large numbers within a few days after patients were treated with them. The experience with erythromycin was better, but the drug was used in six carriers for only ten days. More recently it has been used in a larger number of patients for a longer time and a significant number of resistant strains have appeared. Rather than attempt to eliminate strains resistant to penicillin and tetracycline in our hospitals, it would probably be more effective to avoid use of erythromycin, kanamycin and other agents especially active in staphylococcal infection except in treatment of this infection.

The present studies displayed in a very clear fashion a correlation between susceptibility *in vitro* and susceptibility *in vivo*, and when resistance appeared as measured by *in vitro* tests inhibition no longer occurred *in vivo*. It is our presumption that patients ill with staphylococcal infection will generally respond to antimicrobial treatment as do carriers. When an active drug is given and penetration to the lesion occurs, the staphylococci will be inhibited and healing may result. If resistant strains are present, or develop, then little benefit from treatment will occur.

Some patients with serious staphylococcal infections, however, do not respond well when treated with active agents under optimum conditions. These patients are usually those with impaired resistance to infection from cancer, leukemia, and the like, and failure to respond is a useful although ominous prognostic sign of host failure.

SUMMARY

A study was made of the effect of antibiotics on staphylococci from patients in a mental hospital where drugs had been used infrequently and the staphylococci were largely susceptible.

Erythromycin, to which all cultures before treatment were susceptible, caused a complete suppression of the carrier state, and only a few resistant strains appeared subsequently.

Penicillin, to which 10 per cent of strains were resistant before treatment, caused a drop in the carrier rate to 60 per cent, but the fall was limited at this point by an increase in strains resistant to penicillin.

Tetracycline, to which 4 per cent of strains were resistant before treatment, caused a fall in carrier rates to 20 per cent at which point increase in resistant strains prevented further reduction.

Following treatment, the carrier rates in treated patients increased and the frequency of resistant strains diminished.

Treated non-carriers and non-treated carriers on the same ward did not change during therapy experiments.

Successive treatment of patients with penicillin and tetracycline caused the appearance of a group of cultures resistant both to penicillin and tetracycline, of which a majority did not react with phage (no type). A high proportion of strains of phage type 52–80 susceptible to penicillin and tetracycline initially present in the ward completely disappeared after use of these agents.

Acknowledgment: We wish to acknowledge the help and cooperation of Drs. Oscar Hauk and Lawrence White, of the Central State Hospital, Nashville; Mrs. Joyce McClure and Miss Frances Foster of the laboratory's technical staff.

ADDENDUM

In continued treatment experiments with these patients involving the use of erythromycin, oleandomycin, tetracycline and penicillin, there

has occurred a further shift of phage patterns to 95 per cent group III, largely type 7, 77. A large majority of these cultures are resistant to erythromycin, penicillin and tetracycline.

We have thus duplicated by treatment of hospital patients with antibiotics the situation with respect to the distribution of phage patterns and drug resistance of staphylococci which has appeared in hospitals all over the world following the introduction of antibiotics.

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Recurrent Jaundice of Pregnancy*

A Clinical Study of Twenty-Two Cases

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DURING pregnancy a peculiar type of jaundice may occur, appearing usually late in pregnancy, disappearing soon after delivery, and frequently recurring during subsequent pregnancies. This type of jaundice therefore appears to be directly connected with and provoked by pregnancy. The clinical picture differs from that of other forms of jaundice arising from pregnancy, such as toxemia of pregnancy, and from diseases accompanied by jaundice unrelated to pregnancy, such as hepatitis, choledochus calculi, hemolytic jaundice and exogenic toxic destruction of the liver.

In the earlier literature this type of jaundice received relatively little attention despite the fact that it is not uncommon, according to our experience. Eppinger [*1*] summarized observations in the literature on this pathologic condition, which he called "idiopathic jaundice of pregnancy." Svanborg [*2*] described seven cases of this type of jaundice. Two of these patients had had jaundice during three successive pregnancies, two during two pregnancies. Because of the tendency to recurrence, the name "recurrent jaundice of pregnancy" was suggested. Thorling [*3*] presented a detailed survey of the different types of jaundice during pregnancy. He described thirty-eight cases of this special form of jaundice, which he called "endogenous hepatotoxemia of pregnancy."

The cause of this type of jaundice has not been established. In earlier publications infectious or toxic liver damage was assumed to be present. In investigations with modern liver function tests, however, signs of obstructed bile flow were observed, without any indications of parenchymal liver damage [*2–3*]. In continued studies an additional twenty-two cases have been investigated. The examining procedure has been expanded, and new clinical observations have been made. The results are reported here.

SUBJECTS STUDIED AND METHODS OF STUDY

No systematic investigation of the incidence of jaundice in pregnant women was made. The patients sought medical advice themselves because of symptoms of the disease, or jaundice was observed by the patients' personal physicians during routine checks during pregnancy.

A slight bilirubin increase in the serum seems to occur frequently in the final stage of apparently completely normal pregnancies. The upper limit for physiologic bilirubin concentration in serum in pregnant women is probably higher than in the nonpregnant women [*2–4*]. However, the twenty-two patients included in the present investigation had visible jaundice and a clearly abnormal serum bilirubin increase to more than 2.0 mg./100 ml. The patients ranged in age from twenty to thirty-eight years.

Five patients with pronounced symptoms of the disease were selected for liver biopsy studies. One of them has completed two pregnancies and has had jaundice for four months during each; a biopsy was performed during and after the last pregnancy.

After the pathologic condition had been diagnosed the patients underwent regular examination each week or fortnight during the entire course of the disease. In some of the cases in which the patients had had jaundice during earlier pregnancies regular checkup examinations were made from the third or fourth month of the new pregnancy. Even after the patients' condition appeared clinically to be normal, contact was maintained and renewed examination instituted on the slightest suspicion of liver or biliary disease.

The serum bilirubin concentration was determined according to Jendrassik and Gróf [*5*]. The determination of serum alkaline phosphatase activity was performed according to Buch and Buch [*6*]. The thymol turbidity test according to Maclagan was used [*7*]. The cephalin flocculation test was performed by a modification of Hanger's method [*8*]. Electrophoretic serum protein analysis on paper was carried out according to Goa [*9*]. Proconvertin-prothrombin in plasma was determined by the method of Owren and

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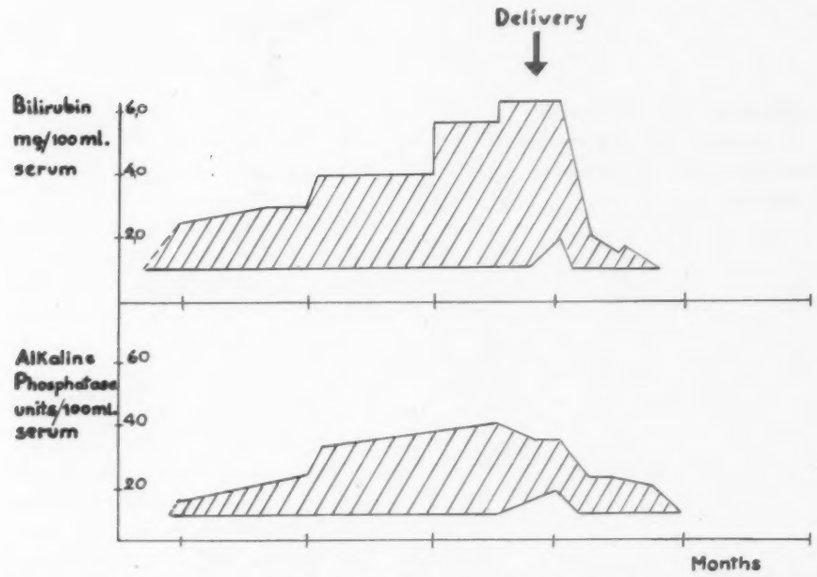

FIG. 1. The serum bilirubin and alkaline phosphatase activity in twenty-two patients during twenty-three pregnancies.

Aas [*10*]. The endogenous formation of carbon monoxide was determined according to a modification of the method of Sjöstrand [*11*] described by Hallberg [*12*].

Roentgenologic investigation of the bile ducts and the gallbladder was carried out after injection of biligrafin (Schering, Germany). Liver biopsy specimens were taken with a Vim-Silverman needle. The patients had fasted ten to twelve hours at the time of examination. The tissue specimens were fixed immediately in 10 per cent formalin, embedded in paraffin, and sections of approximately 5 microns were stained according to van Gieson and with hematoxylin-eosin as well as with Berlin blue. When possible, frozen sections were stained with scarlet red for fat determination.

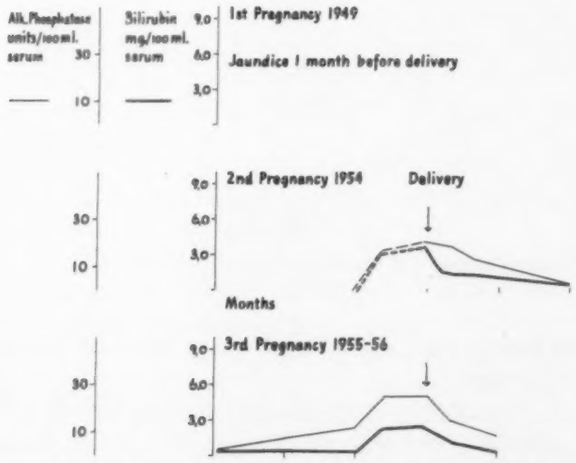

FIG. 2. The serum bilirubin and alkaline phosphatase activity in a patient with jaundice during three pregnancies.

RESULTS

Clinical and Laboratory Studies. In order to establish whether or not hereditary factors determine the occurrence of this disease all patients were questioned in detail as to the incidence of similar conditions in the family. One patient had an older sister who had had a similar pathologic picture during the final stages of two successive pregnancies. A review of the case records from the hospitals where the patient's sister had been treated afforded only an incomplete picture of the course of the disease, but available data definitely indicated that she, too, had, suffered a typical recurrent jaundice of pregnancy. The other twenty-one patients denied knowledge of similar conditions among their relatives.

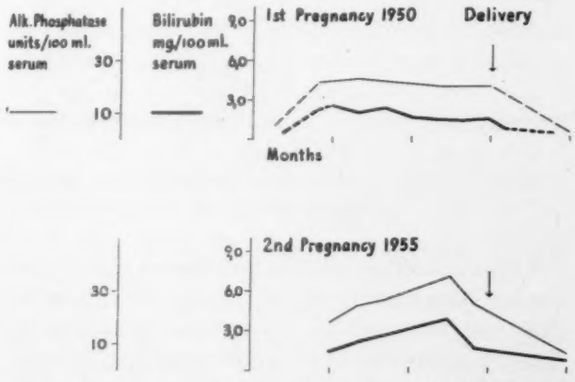

FIG. 3. The serum bilirubin and alkaline phosphatase activity in a patient with jaundice during two pregnancies.

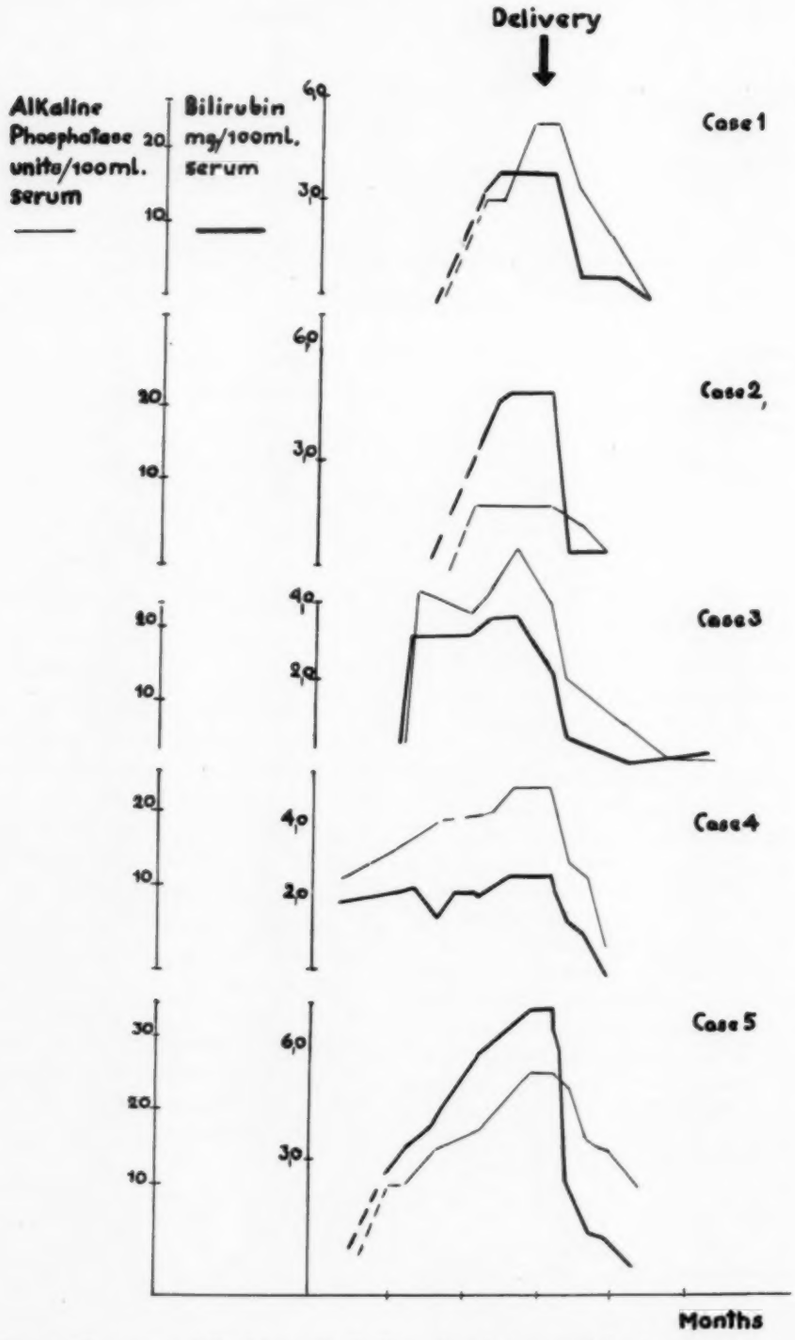

FIG. 4. The serum bilirubin and alkaline phosphatase activity in the patients selected for liver biopsy investigation.

In all cases the onset of the disease was during the last four months of pregnancy. Itching of the skin was reported by seventeen women to be the first symptom. Dark urine, grey discoloration of the faeces, and jaundice were observed within the next few days. In the other five cases itching of the skin was absent. These patients sought medical care only after jaundice had been observed.

During the continued course of the disease the jaundice increased rapidly, as did the itching when present, to reach a maximum after a week or so. The symptoms then persisted at a relatively constant level until delivery. After delivery

the jaundice disappeared first and in all cases within two weeks. As a rule, the itching of the skin persisted about a week longer than the jaundice.

In seventeen patients both jaundice and itching had disappeared within two weeks after delivery. In four the itching persisted another week and in one patient the itching did not disappear until five weeks after delivery.

The patients were also questioned about the development of other symptoms that may occur in hepatic and biliary diseases and in hemolytic forms of jaundice, e.g., fatigue, nausea, vomiting, fever, muscular and joint pains, abdominal pain, diarrhea. Enlargement of the liver or spleen was sought, as were spider angiomas in the skin, enlargement of the lymph glands and the like. The hemoglobin level was determined regularly. In no case did the history or clinical signs indicate hepatitis, cirrhosis of the liver, gallstones or hemolytic jaundice.

All patients were nutritionally in good condition and were little affected by the disease. Many had intensive itching, and scratch excoriations of the skin were observed.

The increase in serum bilirubin was moderate. Parallel with the increase in serum bilirubin, a rise in the serum alkaline phosphatase activity was observed. Together with the clinical symptoms, the serum bilirubin and phosphatase activity increased comparatively rapidly after the onset, and persisted subsequently at a relatively constant level until delivery, whereafter return to normal soon took place. In two cases, however, the serum bilirubin level rose slowly during the entire course of the disease until the time of delivery. The serum alkaline phosphatase activity returned to normal more slowly than the bilirubin level. (Fig. 1.) Examples of individual serum bilirubin and alkaline phosphatase curves from two patients with relapsing jaundice and from the five patients selected for liver biopsy are presented in Figures 2 through 4.

The thymol turbidity test was repeated at all examinations on all patients, and the cephalin flocculation test at all examinations on half the patients. The results were within normal limits and did not change after the disease had subsided. Electrophoretic serum protein investigation was made repeatedly on all patients during and after the periods of disease. No change similar to those that can be found in parenchymal liver disease could be demonstrated.

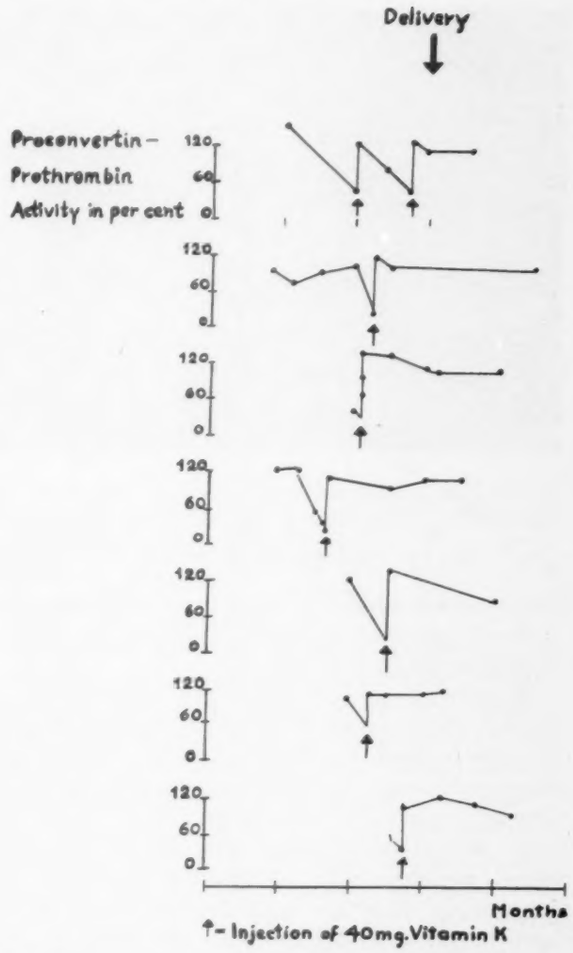

FIG. 5. Proconvertin-prothrombin in serum before and after parenteral administration of vitamin K.

The blood fragility test gave normal values. The von den Bergh reaction was positive, both directly and indirectly. Coombs' test gave negative results. Hemoglobin, serum iron and reticulocyte counts were within limits normal for pregnancy. In two cases the endogenous production of carbon monoxide was measured and found to lie within normal limits, i.e., <0.29 per cent carbon monoxide hemoglobin in the total hemoglobin. Thus, no evidence of a hemolytic origin of the jaundice was obtained.

The proconvertin-prothrombin content in serum was determined at all examinations. In seven cases it dropped to levels threatening a predisposition to hemorrhage. Vitamin K was administered parenterally before complicating hemorrhage occurred. After the administration of 40 mg. vitamin K the proconvertin-prothrombin level returned to normal within one or two days. (Fig. 5.) In one case the proconvertin-

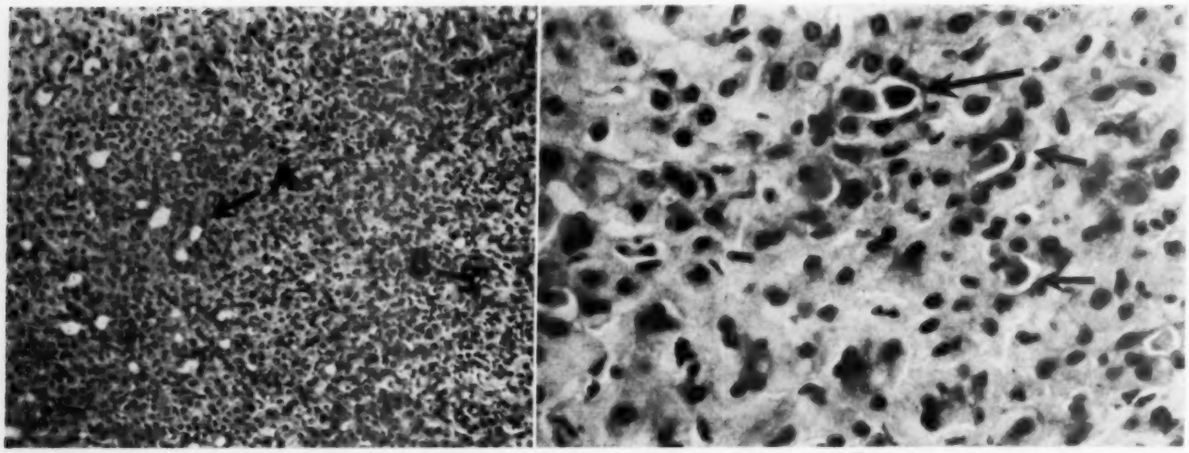

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FIG. 6. Case 1. Low power view of liver tissue showing dilated bile capillaries (A) some with accumulation of bile pigment (B), and occasional associated slight reticular proliferation. van Gieson stain, original magnification × 120.

FIG. 7. Case 1. View showing bile thrombi. van Gieson stain, original magnification × 430.

prothrombin level fell again later in the course of the disease but responded rapidly again to the administration of vitamin K.

Roentgen examination of the bile ducts was carried out in all cases only after disappearance of icterus since satisfactory filling of the bile ducts with contrast medium was not obtained during the period of jaundice. There were no signs of congenital anomalies, biliary stasis of other pathogenesis or residual inability to excrete the contrast medium after the disease had subsided. Four patients had stones in the gallbladder; no concrements were visualized in the bile ducts.

Liver Biopsies. In all tissue specimens the dominant and frequently the only pathologic change observed was an accumulation of bile pigment in the bile capillaries, some of which were greatly dilated. The changes were irregular, focal in distribution and particularly located in the central parts of lobuli, frequently in immediate connection with the central veins. In the liver cells about these bile thrombi the bile pigment was often massed. Many bile capillaries were greatly dilated even when retained bile pigment was not demonstrable in the capillary lumens. Usually no degenerative phenomena were visualized. The liver structure otherwise

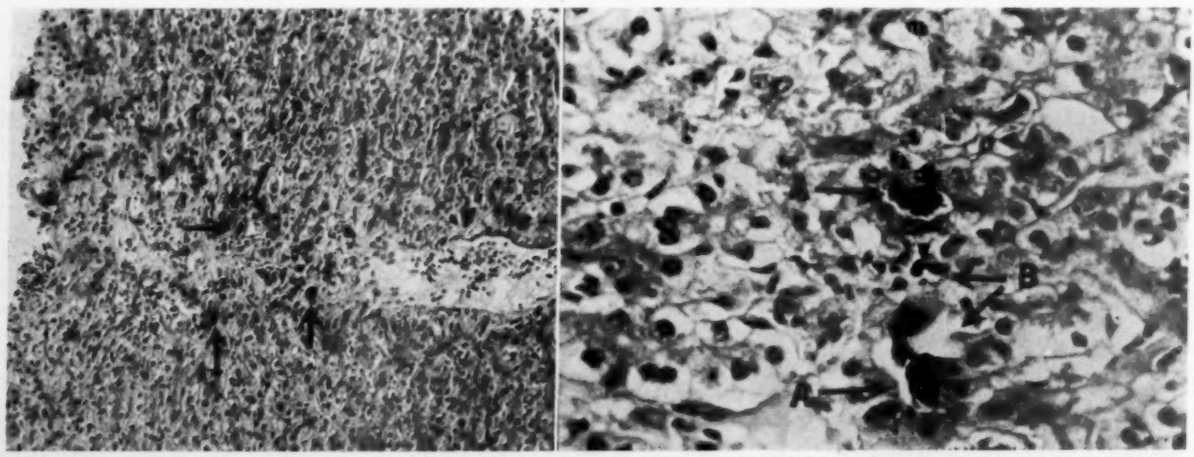

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FIG. 8. Case 2. Section showing many bile thrombi, concentrated especially about the portal region. Reticular proliferation occurs in conjunction with accumulations of bile pigment. van Gieson stain, original magnification × 140.

FIG. 9. Case 2. From the interlobular region. There is an accumulation of bile pigment in dilated bile capillaries (A) and some reticular proliferation in the immediate vicinity, with isolated shrunken, somewhat indistinctly delineated, slightly degenerated hepatic cells (B). van Gieson stain, original magnification × 430.

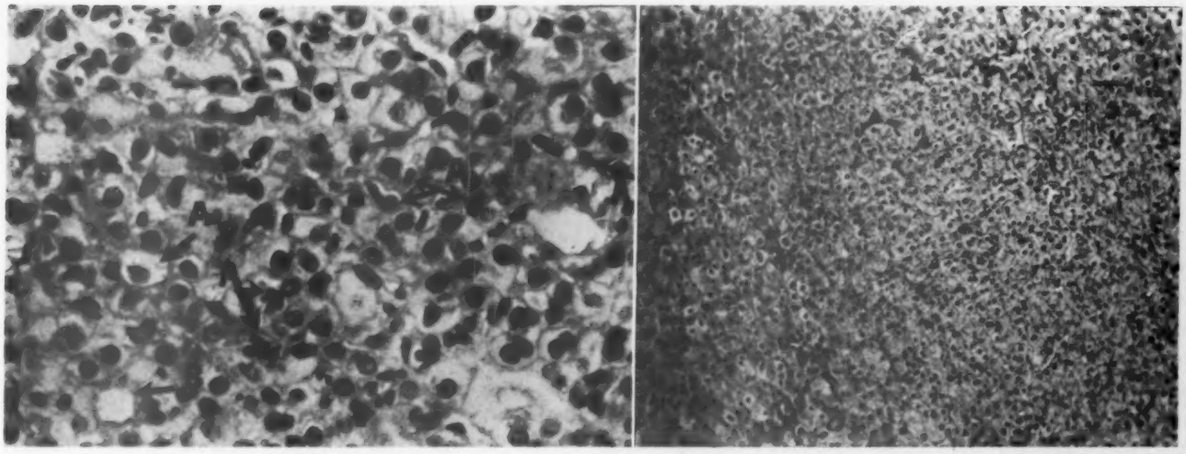

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FIG. 10. Case 3. Section showing many bile thrombi (A) and occasional dilated bile capillaries without bile thrombi (B). Somewhat more markedly reticular than usual in places. van Gieson stain, original magnification × 430.

FIG. 11. Case 3. After delivery. No significant changes, no bile thrombi or dilatation of bile capillaries. van Gieson stain, original magnification × 120.

presented well delimited monomorphic hepatic cells of ordinary size with light and somewhat granular cytoplasm and small monomorphic, homogeneously chromatin-outlined nuclei which appeared to be quite normal.

In association with some of the bile thrombi, especially in the areas with pronounced accumulation of bile pigment, there were isolated hepatic cells exhibiting degenerative phenomena; the cytoplasm was swollen, indistinctly delimited and vacuolized. The nucleus showed irregular chromatin borders and in places was vacuolized. In isolated cells there was a diminished affinity for stain or pyknosis of the nucleus. In the portal connective tissue there was no inflammatory reaction. No dilatation or massing of bile pigment was observed in the bile ducts in the portal regions or in the intralobular ductuli. In one case (Case 1, Fig. 4) there were areas with especially marked accumulation of bile pigment associated with round cells and isolated leukocytes as well as reticular proliferation. In the regions with less pronounced accumulation of bile pigment and in the areas without signs of biliary stasis no degenerative changes were observed in the liver cells. Special staining did not reveal hemosiderin in the liver cells. In the two cases investigated after delivery no residual changes were demonstrated in the liver, which had a completely normal structure.

The characteristic biopsy observations are presented in Figures 6 through 13.

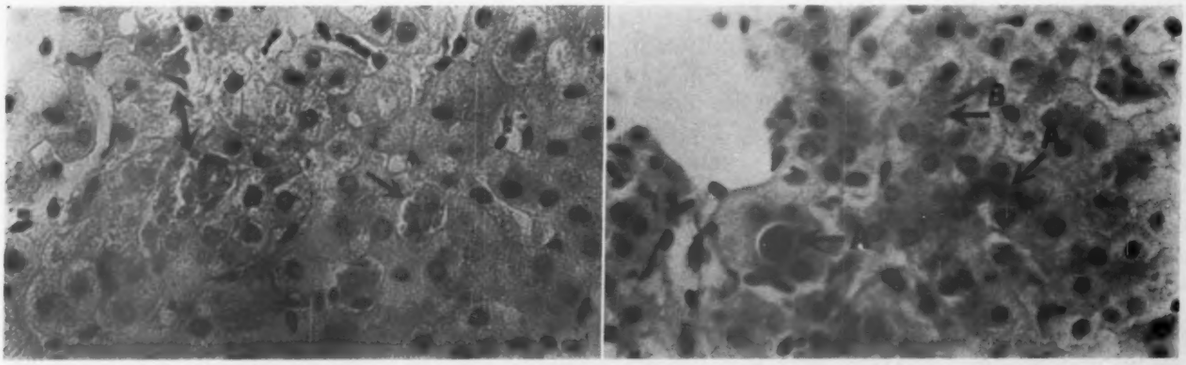

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FIG. 12. Case 4. Liver with isolated, porous, pale bile thrombi; no other pathologic changes. van Gieson stain, original magnification × 520.

FIG. 13. Case 5. Liver with bile thrombi (A) and accumulation of bile pigment in hepatic cells (B). van Gieson stain, original magnification × 520.

COMMENTS

The symptoms were relatively mild in all cases, never of such severity that the general condition of the patients deteriorated. The dominating subjective symptom was itching of the skin; mucosal itching was not reported. This itching was so severe in some cases that it disturbed the patient's sleep at night. Symptoms of hepatitis, such as anorexia, fever, muscle and joint pains, vomiting, enlargement or tenderness of the liver, and the like were absent. Pain, symptoms of cholangitis and dyspepsia and other complaints commonly accompanying gallstones also were absent. Nor were there signs indicating a hemolytic origin of the jaundice. The itching of the skin observed in most of these cases does not occur in the hemolytic forms of jaundice. In order to eliminate definitely the possibility of a hemolytic pathogenesis the endogenous carbon monoxide production was analyzed; in hemolytic states the production of carbon monoxide is increased. The results in these cases were within normal limits, which could not have been the case in hemolytic jaundice of the same degree of severity. This precludes the possibility of a hemolytic origin of this type of jaundice.

It has been established beyond doubt that this disease has a direct connection with pregnancy. The patients had no similar symptoms during the intervals between pregnancies. In all cases rapid improvement followed delivery.

A further confirmation of the relationship to pregnancy is the tendency to recur in subsequent pregnancies. There was recurrence in four of six women who had undergone more than one full-term pregnancy; in one of them during three successive pregnancies and in three during two successive pregnancies. The other two had previously had normal pregnancies without demonstrated jaundice. One of these women had visible jaundice without itching of the skin for the first time during her second pregnancy, the other initially during her third pregnancy. Previously [*2*] two women with recurrences during three pregnancies and two women with two recurrences were recorded. We have not seen cases in which pregnancy with jaundice was followed by pregnancy without jaundice. It therefore seems that the disease need not occur during all pregnancies but the risk of recurrence is great if it has once occurred. This observation does not agree, however, with the results of Thorling's investigations [*3*]. He found five cases in which the patients were completely healthy during a later pregnancy despite earlier pregnancy with jaundice. Three patients with jaundice in earlier pregnancy had only itching of the skin in later pregnancy.

Another striking feature is the similarity of the course in the individual case in repeated pregnancies. In all cases the onset of the symptoms occurred at the same stage as in prior pregnancies. Among the women observed with jaundice during three pregnancies, symptoms appeared in two women two months before delivery in all three pregnancies, and in one case three months before delivery. In two of these cases the time of onset could be stated rather exactly as regular examinations were carried out during the greater part of the pregnancy. Even the rate at which the symptoms disappear after delivery seems to be subject to individual variation. This is illustrated especially by one case in which the patient had symptoms four months before delivery and itching of the skin persisting unusually long, i.e., five weeks after delivery, in conjunction with two pregnancies. The course of the disease in one patient with three recurrences and in one with two recurrences is shown in Figures 2 and 3.

The cause of this disease is not known. The clinical picture and the course of the disease are consistent with those occurring in cases of biliary stasis. Also indicative of this possibility are the results of liver function tests. The parallel increase of serum bilirubin and alkaline phosphatase activity and the absence of signs of parenchymal liver damage have been pointed out previously [*2*]. Thorling [*3*] also found a greater increase in the serum alkaline phosphatase activity in these women with jaundice than is found in pregnancy without this type of jaundice.

The fall in the proconvertin-prothrombin level observed in a number of these patients and the rapid return to normal after parenteral administration of vitamin K definitely indicate the presence of biliary stasis and not of significant parenchymal liver damage. In parenchymal liver disease with jaundice of a corresponding degree of severity, with a decline in the proconvertin-prothrombin level, such a rapid return to normal following administration of vitamin K does not occur.

Similar types of jaundice have been described as a complication following the administration of chlorpromazine, methyltestosterone and Salvarsan.® The patients in the present series

received no such therapy. They were on a conventional diet and lived under satisfactory hygienic conditions. Exposure to any substance injurious to the liver could not be demonstrated.

One of the twenty-two patients had a sister who in all probability suffered from the same disease. All other patients definitely denied having any knowledge of a similar disease in relatives. These relatives, however, were questioned by the patients and not by ourselves, and from this point of view this investigation has not been carried out with sufficient accuracy to permit definite conclusions concerning any hereditary predisposition to this disease.

There have been earlier histologic liver studies on pregnant women (mainly on postmortem material) in circumstances in which the pregnancy has been complicated by fatal, frequently infectious diseases. On the basis of these pathologico-anatomic investigations it has been concluded that the liver is altered in pregnancy. Hepatic cell damage, e.g., by fatty degeneration, has been described [*13*] and designated "pregnancy liver." In later investigations of the liver no characteristic changes of the liver in pregnancy have been demonstrated. In a study of liver biopsy material, in which postmortem tissue changes are precluded, Ingerslev and Teilum [*14–15*] and Nixon et al. [*16*] demonstrated that there are no characteristic histological changes in normal pregnancy. A certain degree of polymorphism of the cell nuclei, polynuclear hepatic cells, slight fatty deposition and, in isolated cases, reactive changes with lymphocyte infiltration may occur. Nixon et al. [*16*] also investigated liver biopsy material from patients with jaundice during pregnancy. In the majority of cases hepatitic changes of varying degree were demonstrated. In two cases biliary stasis was noted, without signs of hepatitis. The incomplete presentation of clinical data makes it impossible, however, to differentiate cases of clinically typical hepatitis from possible cases of jaundice of the recurrent jaundice of pregnancy type.

According to a report by Ljunggren [*17*], Welin observed biliary stasis and bile thrombi in five of six cases of jaundice during pregnancy. In two of the five cases he also found slight degenerative hepatic cell changes. Detailed clinical data relating Ljunggren's biopsy findings are not given.

In the cases reported herein the picture was dominated completely by biliary stasis with dilatation of the bile ducts and precipitation of bile pigment ("bile thrombi"), whereas hepatic cell degenerative phenomena were inconstant and usually of negligible importance. These degenerative changes occurred principally in or in the immediate vicinity of the hepatic cells bordering upon capillaries filled with bile thrombi. No degenerative phenomena were observed around the remainder of the dilated bile capillaries or in the remainder of the liver parenchyma. The slight signs of degeneration may therefore surely be regarded as secondary to the biliary stasis. Such changes are observed in cases of biliary stasis due to obstruction of the common bile duct. The insignificant reactive changes would also seem to be explicable as secondary to the biliary stasis. The histologic picture accordingly offered no ground for the assumption of primary inflammation or degeneration of the liver parenchyma ("infectious or toxic liver damage"). In cases of obstruction of the common duct there is often dilatation of the bile ducts in the portal region. Such changes did not occur in the present biopsy material nor was there definite dilatation of intralobular ductuli. The extremely limited number of portal regions in the biopsy material, however, did not permit definite exclusion of changes in the larger bile ducts. Thus, on the basis of these biopsy investigations it is not possible to express an opinion concerning the site of the immediate cause of the changes in bile flow. The symptom picture is consistent with an obstruction to the bile flow of either intrahepatic or extrahepatic origin. On the basis of the existing facts only speculation, from which we shall refrain, is as yet possible on this question.

There is a tendency to classify the complications of pregnancy with uncertain pathogenesis together under the heading of toxemia of pregnancy. Thorling [*3*] has entitled this pathologic condition "endogenous hepatotoxaemia of pregnancy." As long as the pathogenesis is unknown, the nomenclature will remain variable. Hepatic lesions occur in toxemia of pregnancy. In advanced stages, liver necrosis and jaundice may appear. In these cases, however, the jaundice is a relatively inconsequential symptom in a grave disease picture that frequently involves the threat of death for both mother and fetus. As far as we are able to determine, recurrent jaundice of pregnancy differs in its clinical picture, course and prognosis from those accompanying the hepatic injury during

toxemic conditions. It therefore seems important to attempt to distinguish between these two pathologic conditions in nomenclature as well as in other aspects. As mentioned earlier, no signs of liver necrosis or diffuse parenchymal liver lesions were demonstrated in any of these cases. One patient had concurrent proteinuria without a rise in blood pressure during the last two weeks of pregnancy. This proteinuria was designated nephropathia gravidarum by the patient's gynecologist. The other patients did not have symptoms suggestive of toxemia. The absence of concurrent symptoms of toxemia further indicates that the jaundice herein described is not a manifestation of the toxemia of pregnancy.

It is essential that the tendency of the prothrombin level to decline in these patients be observed and counteracted. Seven patients had a decrease in prothrombin that in all probability would have caused an increased tendency to hemorrhage if vitamin K had not been given parenterally. Thorling [*3*] mentions three cases of jaundice in which the fall in the prothrombin index and jaundice were first observed at delivery and in which there was an increased tendency to bleeding.

The prognosis for the mother seems to be good. Signs of residual liver disease or biliary stasis could not be demonstrated in any case. The three patients described herein and previously [*2*] who had jaundice during three successive pregnancies are still apparently in good health five and one-half, five and two years after the last period of jaundice. The absence of histologically demonstrable liver changes in the two cases investigated after delivery support this prognostic assumption. One of them (Case 3, Fig. 4) had completed two pregnancies with jaundice during four months of each pregnancy.

The prognosis seems to be good for the child as well. One died during delivery because of rupture of the tentorium cerebelli. There was no reason to regard this complication as associated with the mother's disease. All other infants in this series appeared to be in normal health.

The disease seems to be considerably more common, at least here in Sweden, than the sparse reports in the literature indicate. No definite data on incidence are as yet available. The cases reported herein constituted 0.2 per cent of the total number of deliveries during the period covered by the investigation. The disease must certainly occur more frequently.

In an earlier study [*2*] the descriptive name "recurrent jaundice of pregnancy" was selected in consideration of the obvious tendency to recurrence. Final designation of this pathologic condition must wait until the pathogenesis has been further clarified.

SUMMARY

Clinical observations and the results of liver function tests in twenty-two patients with what we have designated recurrent jaundice of pregnancy are reported. The disorder differs clinically and in other manifestations from other types of jaundice occurring during pregnancy.

Seven patients showed a drop in the proconvertin-prothrombin levels of the serum and a potential general tendency to bleeding. If this complication can be diagnosed in time, hemorrhage can be prevented by the parenteral administration of vitamin K, which results in a rapid return to normal of the proconvertin-prothrombin level.

All patients who had had jaundice during a prior pregnancy had a recurrence of the disease in a subsequent pregnancy. The clinical picture durring the recurrence was strikingly similar to the previous attack.

Liver biopsy was performed during periods of jaundice in five patients. The histologic observations were dominated by signs of biliary stasis with dilatation of the bile ducts and accumulation of masses of bile pigment. No residual liver changes were observed in two patients investigated again after delivery and after the clinical picture had become normal.

The prognosis appears to be good; residual symptoms have not been demonstrated even after three recurrences.

The cause of the disease seems to be impairment of bile flow. The pathogenesis of this biliary stasis is not known.

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Biochemical, Blood Gas and Peripheral Circulatory Alterations in Hepatic Coma*

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THE neuropsychiatric features of hepatic coma have been extensively reviewed [*1–4*]. Additional clinical features exhibited by such patients are hyperpnea [*5,6*], arterial oxygen unsaturation [*7,8*], peripheral vasodilatation [*9,10*], and oliguria with azotemia [*11*]. These alterations may influence the neurological state, and could account in part for the reported poor correlation between blood ammonia concentrations and level of consciousness [*12–15*]. They may also provide other diagnostic parameters for the recognition of occult cerebral dysfunction and the prognosis of frank coma.

It is the purpose of the present study to describe observations on the arterial pH and CO_2 tension, arterial oxygen saturation, peripheral arterial-venous oxygen difference and blood urea nitrogen in patients with disease of the liver with and without hepatic coma. In particular, these values have been related to level of consciousness, arterial ammonia concentration and peripheral arterial-venous ammonia difference in groups of patients and in the same person studied serially through one or more episodes of coma.

MATERIALS AND METHOD

Forty-nine patients with liver disease were studied; all were hospitalized men ranging in age from twenty-eight to seventy years. The diagnosis was usually apparent clinically; histologic confirmation from biopsy or autopsy specimens was obtained in forty patients. Thirty-six patients had alcoholic nutritional cirrhosis; six of these had "florid" cirrhosis [*16*], and two had hepatomas. Nine patients had hepatitis which was chronic in six and fulminant in three. Two patients had postnecrotic cirrhosis; one had hemachromatosis and another congestive heart failure.

Coma was graded immediately prior to each sampling time as follows: grade 0, no abnormality detected, twelve patients; grade 1, deficiencies in speech, orientation, memory, judgment or calculation apparent with detailed examination; grade 2, deficiencies listed in grade 1 but apparent with less detailed examination; grade 3, stuporous; grade 4, semicomatose or comatose. For statistical correlation of group data, grades 1 and 2 and grades 3 and 4 were combined; seven patients and thirty patients, respectively. These patients were studied during forty-six separate admissions (at least three months between discharge and readmission), constituting a total of twelve episodes of grades 1 to 2 and thirty-four episodes of grades 3 to 4.

Multiple factors precipitated coma in this study. Profound liver failure was the most common cause, occurring in eighteen instances; in four patients hemorrhage occurred immediately prior to death. Other precipitating factors included an oral nitrogenous load in eleven episodes, and gastrointestinal hemorrhage in twelve episodes; each appeared in patients with well developed portal collaterals and minimal-to-marked liver dysfunction. Pyogenic infection and abdominal paracentesis each precipitated coma in two instances, and chlorothiazide was a questionable precipitating factor in one.

Treatment in patients graded 1 to 4 was primarily directed at reduction of the blood ammonia [*17,18*]. All nitrogenous substances (protein, drugs, etc.) were initially eliminated from the diet. Protein in increments of 20 gm. was subsequently given, dependent upon the patient's progress. The bowel was emptied and "sterilized" with neomycin [*19,20*] or tetracycline [*18,21*]. At least 1,600 calories were given as hypertonic glucose solution. Patients with hepatitis and "florid" cirrhosis received steroids. The details of this regimen will be published elsewhere [*22*].

Fourteen patients were studied serially during episodes of coma. Five of the seventeen patients who died were studied within twenty-four hours before death.

Unless specified, all determinations were made on arterial blood obtained from a brachial artery. When the arterial-venous (A-V) difference was determined, venous blood (withdrawn without stasis fifteen minutes after adjustment to needles) was obtained from a medial subcutaneous antecubital vein. Serial venous determinations were obtained from the same vein.

* From the Department of Medicine, Duke University School of Medicine and Veterans Administration Hospital, Durham, North Carolina. Supported in part by a grant from the American Heart Association.

Blood ammonia (NH_3) was measured by a modification of the microdiffusion method of Brown et al. [*23,24*]. The pH of whole blood was measured with a Cambridge Model R pH meter equipped with an enclosed glass electrode. Measurements were made at room temperature, usually 25° to 26°C., and the results corrected to 37°C. by Rosenthal's factor [*25*]. The CO_2 content of whole blood was determined by the method of Van Slyke and Neill [*26*]. The CO_2 tension was calculated from this value, the pH and the hemoglobin concentration by the line chart of Van Slyke and Sendroy, using a pK of 6.11 [*27*]. Oxygen content and saturation were determined by the spectrophotometric method of Hickam and Frayser [*28*]. The urea nitrogen of the blood was measured by Conway's microdiffusion method [*29*].

RESULTS

Blood Ammonia in Relation to Level of Consciousness. Arterial and venous ammonia concentrations were compared in patients with liver disease with and without hepatic coma. (Fig. 1.) The mean arterial ammonia concentration for patients with grade 0 was significantly higher than that of normal control subjects, $p = <0.01$. When group means for arterial ammonia concentration were compared in patients with liver disease (grades: 0 = 132 μg./100 ml., 1 to 2 = 174 μg./100 ml., 3 to 4 = 268 μg./100 ml.) a significant increase was evident between each of the groups, $p = <0.01$. The mean arterial ammonia readings were similar to their corresponding venous values in patients with grade 0 but were significantly greater with altered consciousness; $p = <0.01$ and 0.02 for grades 1 to 2 and 3 to 4, respectively.

Overlap of arterial ammonia concentrations was evident with all grades of consciousness. (Fig. 1.) Arterial values above 160 μg./100 ml. were obtained in one patient graded as 0 (W. F.). (Table 1.) Six patients graded 1 to 2 had arterial concentrations above 200 μg./100 ml., and one (W. F.) had a value above 300 μg./100 ml.; 16 per cent of observations were below the arterial mean for grade 0, but none were below 112 μg./100 ml. Arterial readings below 153 μg./100 ml. were not encountered in any patient with grades 3 to 4.

Serial arterial and venous concentrations obtained during one or more episodes of coma in the same patient are shown in Table 1. Correlation between arterial ammonia concentration and level of consciousness was good in ten patients; in four (F. C., L. P., R. S., V. C.) there was occasional overlap between grades 1 to 2 and 3 to 4. In all six patients studied when coma was absent arterial readings were well below values obtained during coma. Gross discrepancies in correlation between venous ammonia concentrations and level of consciousness were evident in three patients (L. McN., J. B., R. S.).

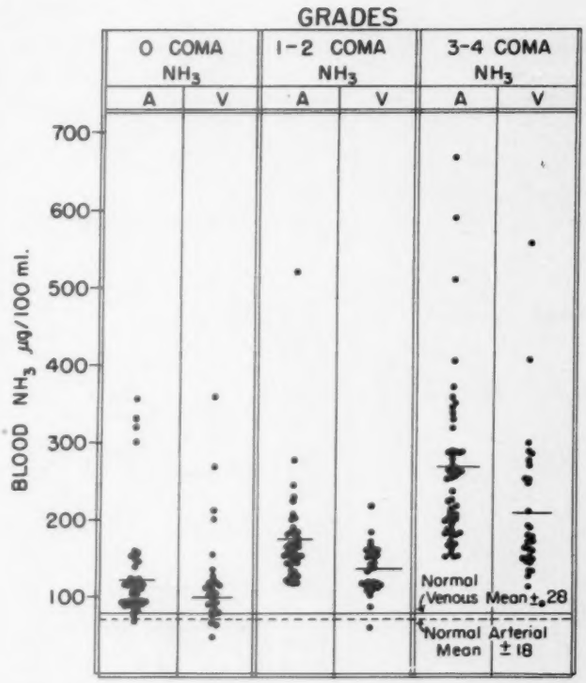

FIG. 1. Arterial (135 observations in forty-nine patients) and peripheral venous (ninety-two observations in thirty-five patients) ammonia concentrations in patients with liver disease at three levels of consciousness. Horizontal lines represent mean values for each group.

Peripheral A-V Ammonia Difference in Relation to Arterial Ammonia Concentration and Peripheral A-V Oxygen Difference. In these patients with liver disease peripheral A-V ammonia differences were positive, with four exceptions (V. C., T. L., J. W., F. C.—Table 1), and correlated with arterial ammonia concentration. (Fig. 2.) The correlation coefficient for these data was significant ($r = 0.43$, $p = <0.01$). A positive A-V difference of more than 100 μg./100 ml. was obtained only with arterial values above 200 μg./100 ml. Serial determinations in individual subjects (Table 1) did not show a consistent relationship between these parameters.

A comparison of the mean peripheral A-V oxygen differences in ten normal subjects ($\bar{d} = 4.27$ volumes per cent) with that of twenty-three patients with liver disease ($\bar{d} = 2.16$ volumes per cent), grades 0 to 4, revealed a significant decrease in the latter group, p =

TABLE 1

BIOCHEMICAL AND BLOOD GAS DETERMINATIONS IN PATIENTS WITH LIVER DISEASE MEASURED SERIALLY DURING ONE OR MORE EPISODES OF COMA

Patient	Diagnosis	Date	Coma		NH_3 (μg. %)			O_2		pH	CO_2 (mM./L.)	pCO_2 (mm. Hg)	Blood Urea N (mg. %)
			Precipitating Factors	Grade	Arterial	Venous	A-V	Arterial (% saturation)	A-V (volumes %)				
L. McN.	Cirrhosis*	5-17-56	High protein	4	270	211	59	....	....	....	....	....	....
		5-18-56	.....	3	225	114	111	....	....	....	....	....	....
		5-21-56	.....	0	80	76	6	....	....	....	....	....	....
R. S.	Cirrhosis*	5-21-56	.....	0	111	93	18	....	....	....	....	....	....
		5-24-56	Abdominal paracenteses	2	203	144	59	....	....	7.46	....	....	....
		6-6-56	Septicemia	2	176	103	73	....	....	....	....	....	....
		6-18-56	.....	0	109	90	19	....	....	....	....	....	....
		7-17-56	Septicemia	1	145	98	47	94.3	....	7.48	21.1	37.0	....
		7-23-56	.....	1	156	122	34	94.1	....	7.44	21.6	36.0	....
		8-28-56	Hemorrhage	3	214	92	122	....	....	....	....	....	....
		11-6-56	Hemorrhage, mild to	1	156	135	21	....	....	....	....	....	....
		11-8-56	marked	1	153	109	44	....	....	....	....	....	6.2
		12-4-56†		3	194	...	...	88.0	....	7.39	22.1	41.5	....
S. T.	Cirrhosis*	5-31-56	Hemorrhage	4	253	189	64	....	....	....	....	....	....
		6-6-56	.....	1	133	117	16	....	....	....	....	....	....
		6-25-56	.....	1	119	118	8	....	....	....	....	....	....
		4-29-57	Hemorrhage	2	172	150	22	....	....	....	....	....	....
G. S.	Cirrhosis*	7-12-56	Liver failure	1	122	118	4	95.9	....	7.52	22.7	33.5	....
		7-17-56	.....	1	131	92	39	94.8	....	7.44	19.5	34.5	36.6
		8-2-56	Hemorrhage	4	590	558	32	96.6	....	7.55	13.8	17.9	....
V. C.	Cirrhosis*	10-2-56	Hemorrhage	1	151	116	35	....	....	....	....	....	16.7
		10-26-56	Hemorrhage plus sur-	2	170	177	−7	89.9	....	7.46	22.4	37.0	....
		10-29-56	gery	0	81	65	16	84.0	....	7.46	23.9	39.1	39.8
		12-4-56	.....	3	224	...	...	89.1	....	7.42	24.3	38.0	....
		12-11-56	Hemorrhage	4	359	300	59	....	....	....	....	....	....
		12-14-56	.....	1	160	160	0	88.3	....	7.46	23.7	37.1	10.3
		12-20-56	.....	2	209	161	48	....	....	....	....	....	12.6
		9-6-57	Liver failure	3	181	...	...	88.4	....	7.46	24.7	38.6	....
T. L.	Cirrhosis,* hepatoma	1-15-57	Liver failure	1	144	117	27	92.9	1.24	7.42	17.0	31.0	17.6
		1-23-57	.....	1	144	149	−5	....	1.73	....	....	....	14.8
		2-18-57	Hemorrhage	3	201	171	30	91.7	2.54	7.44	16.4	27.6	61.6
		2-19-57	.....	3	252	226	26	92.1	1.13	7.42	14.9	26.1	70.2
		2-20-57†	.....	4	345	252	93	84.8	3.40	7.16	13.5	42.3	73.6
J B.	Cirrhosis*	1-23-57	Hemorrhage	4	350	135	215	92.3	6.05	7.51	15.1	22.4	18.0
		1-24-57	.....	4	218	147	71	....	4.98	7.45	13.4	22.9	....
		1-25-57	.....	1	169	87	82	94.5	4.97	7.41	14.4	26.0	18.1
		2-14-57	High protein	2	184	136	48	93.0	3.83	7.45	21.0	34.4	10.5
		2-20-57	.....	3	288	247	41	93.5	6.40	7.46	16.5	26.7	9.2
		3-4-57	.....	0	85	81	4	92.4	4.59	7.43	20.6	35.4	4.6
J. W.	Fulminant hepatitis	2-1-57	Liver failure	4	258	262	−4	89.3	1.18	7.46	24.5	43.0	26.7
		2-4-57	.....	4	406	408	−2	....	1.70	7.28	22.5	54.3	89.6
W. F.	Chronic hepatitis, postoperative portocaval shunt	3-6-57	.....	0	320	...	...	97.2	....	7.43	20.4	36.1	14.9
		7-18-57	.....	0	300	213	97	....	....	....	....	....	12.1
		10-10-57	.....	0	...	359	...	....	....	....	....	....	11.7
		12-10-57	.....	0	...	269	...	....	....	....	....	....	....
		2-4-58	.....	0	332	201	131	....	....	7.39	21.0	40.9	....
		2-25-58	.....	0	357	...	...	....	....	....	....	....	....
		3-20-58	? Chlorothiazide	2	520	...	...	....	....	....	....	....	26.8
		3-27-58	.....	0	200	...	...	....	....	....	....	....	....
		4-9-58	.....	0	167	...	...	....	....	....	....	....	....

*Alcoholic-nutritional.

†Observations made within twenty-four hours before death.

TABLE 1 (*Continued*)

BIOCHEMICAL AND BLOOD GAS DETERMINATIONS IN PATIENTS WITH LIVER DISEASE MEASURED SERIALLY DURING ONE OR MORE EPISODES OF COMA

Patient	Diagnosis	Date	Coma		NH_3 (μg. %)			O_2		pH	CO_2 (mM./L.)	pCO_2 (mm. Hg)	Blood Urea N (mg. %)
			Precipitating Factors	Grade	Arterial	Venous	A-V	Arterial (% saturation)	A-V (volumes %)				
F. C.	Cirrhosis,* post-operative spleno-renal shunt	4-1-57	Hemorrhage plus surgery	3	180	180	0	81.2	0.09	7.44	14.6	25.0	27.1
		4-2-57		4	331	...	...	90.0	....	7.38	14.1	27.6	52.1
		4-3-57	.....	4	277	...	...	....	....	....	....	....	38.1
		4-4-57	.....	3	224	...	...	....	....	....	....	....	27.5
		4-5-57	.....	3	200	193	7	90.0	1.08	7.46	15.7	26.3	22.5
		4-8-57	.....	1	186	184	2	89.6	0.65	7.40	15.5	29.2	14.6
		4-17-57	Hemorrhage	3	264	...	...	....	....	7.47	18.9	30.3	17.9
		4-18-57	.....	3	252	253	−1	....	0.93	....	....	....	14.1
		4-19-57	.....	1	176	...	...	....	....	....	....	....	17.9
W. B.	Cirrhosis*	9-17-57	High protein plus Diamox®	3	290	...	...	95.7	....	7.34	7.6	14.2	23.0
		9-19-57		4	289	...	...	94.0	....	7.42	14.7	22.9	17.1
		9-20-57	.....	4	287	286	1	93.5	.81	7.42	11.2	17.4	.....
		9-23-57	.....	2	165	163	2	91.2	.37	7.42	14.3	24.3	41.3
		9-26-57	.....	1	163	154	9	93.6	.36	7.34	15.0	30.3	45.6
L. P.	Chronic hepatitis	11-13-57	High protein plus liver failure	3	319	148	171	95.5	....	7.39	20.1	39.1	8.4
		11-15-57		2	245	...	...	94.8	....	7.38	20.2	40.0	.....
		11-18-57	.....	0	89	...	...	....	....	....	....	....	10.3
		1-2-58	Liver failure	3	284	...	...	....	....	....	....	....	9.4
		1-3-58	.....	3	254	...	...	96.1	....	7.39	21.2	41.1	.....
		1-6-58	.....	3	238	...	...	93.1	....	7.40	22.5	43.5	8.2
		1-13-58	.....	2	192	134	58	....	....	....	....	....	8.0
		1-14-58	.....	2	188	115	63	....	....	....	....	....	11.8
		1-20-58	.....	4	510	...	...	87.1	....	7.52	21.4	32.5	5.8
		1-21-58	.....	4	201	...	...	78.5	....	7.43	24.5	43.6	6.6
		1-22-58†	.....	4	427	...	...	91.8	....	7.58	25.1	31.8	6.6
W. H.	Cirrhosis*	11-27-57	Hemorrhage	4	371	...	...	88.2	....	7.56	21.6	29.2	50.5
		11-29-57	.....	4	425	...	...	....	....	7.49	21.7	....	71.3
		12-2-57†	.....	4	668	...	...	88.8	....	7.34	13.0	28.0	129.0
H. R.	Cirrhosis,* "florid"	1-20-58	Liver failure	3	158	134	24	90.6	2.02	7.41	16.6	30.5	26.0
		1-21-58	.....	3	168	...	...	92.9	....	7.44	18.6	32.3	33.0
		1-22-58	.....	3	160	...	...	89.0	....	7.44	19.0	32.4	35.0
		1-27-58	.....	1	112	...	...	92.6	....	7.40	16.5	30.8	34.9

* Alcoholic-nutritional.
† Observations made within twenty-four hours before death.

<0.01. Serial determinations of A-V oxygen differences in five patients during one or more episodes of coma (Table 1) showed occasional gross fluctuations, which in one instance (T. L.) was coincident with terminal hypoxia and shock and in another (J. B.) appeared randomly. In three patients (W. B., J. W., E. C.) values remained fairly constant despite considerable alteration in other measured parameters and in level of consciousness.

When A-V ammonia differences were compared with A-V oxygen differences (Fig. 3), a proportionate relationship was observed, $r = 0.69$, $p = <0.01$. Peripheral ammonia A-V difference was minimal and not greater than 15 μg./100 ml. when the A-V oxygen difference was below 1.0 volume per cent, despite the degree of arterial ammonia concentration. With A-V oxygen differences between 1.0 and 2.0 volumes per cent, A-V ammonia differences were variable, but never greater than 30 μg./100 ml. However, A-V oxygen values above 2.0 volumes per cent were accompanied by peripheral A-V ammonia differences of at least 15 μg./100 ml. when arterial ammonia concentrations were above 158 μg./100 ml.

Arterial pH and CO_2 Tension in Relation to Level of Consciousness. Arterial pH was correlated with CO_2 content in twenty-three patients. (Fig. 4.) When group means for pH (grades: 0 = 7.42, 1 to 2 = 7.44, 3 to 4 = 7.42) were compared with the mean arterial pH (7.38) of eighteen normal subjects, comparable in age, each was significantly greater; $p = <0.01$ for the former two groups and <0.05 for the latter more cerebrally depressed patients. The mean pCO_2

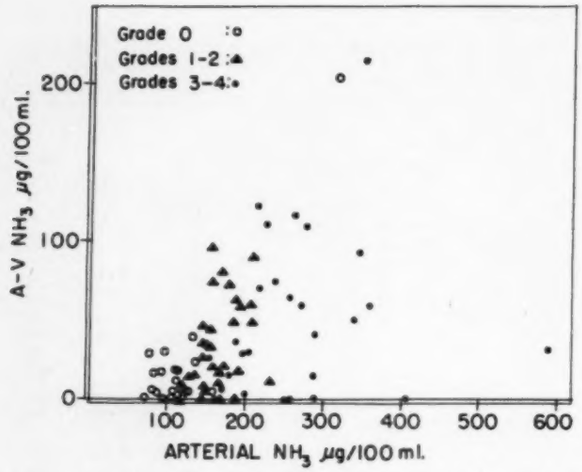

FIG. 2. Peripheral arterial-venous ammonia differences in patients with liver disease at three levels of consciousness (ninety-two observations in thirty-five patients).

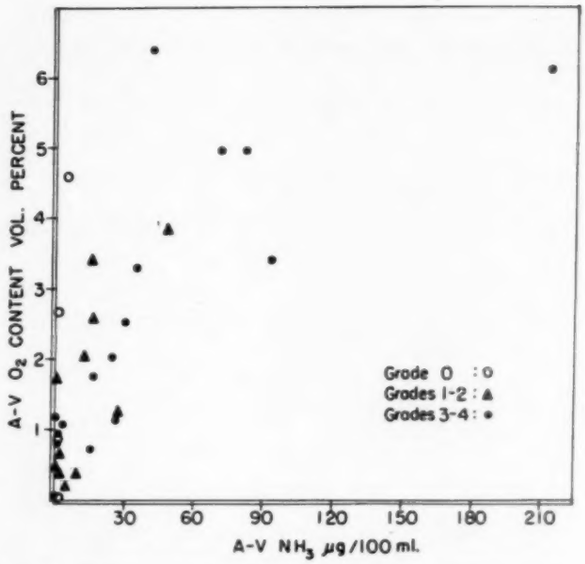

FIG. 3. Peripheral arterial-venous ammonia difference in relation to peripheral arterial-venous oxygen difference in patients with liver disease at three levels of consciousness (thirty-two observations in eighteen patients).

of all patients with liver disease (grades: 0 = 37.5 mm. Hg, 1 to 2 = 33.3 mm. Hg, 3 to 4 = 31.8 mm. Hg) was significantly lower ($P = <0.01$) than that calculated for controls (mean = 44.6 mm. Hg). The difference between the grade 0 group and those with coma graded 1 to 4 was also significant ($p = <0.01$).

Correlation between arterial pH and level of consciousness was generally good when individual subjects were studied during one or more episodes of coma. (Table 1.) In four patients (W. B., H. R., J. B., L. P.) the values appeared to rise in early coma, attain a relatively fixed level unless coma was progressive, and fall towards normal, with slight delay, with increasing consciousness. The lowest pH levels were seen in three patients (T. L., J. W., W. H.) studied within two to fourteen hours prior to death, and one (W. B.) who was severely acidotic prior to the development of coma. A sudden fall in pH during grade 4 coma, not related to imminent death, was seen in two patients (F. C., L. P.); in the former this was coincident with recovery from anesthesia and in the latter with an obstructed airway.

Arterial Oxygen Saturation in Relation to Level of Consciousness and pCO_2. Arterial oxygen saturation was measured in twenty-three patients (fifty-eight observations). Although mean concentrations decreased slightly with coma (grades: 0 = 93 per cent, 1 to 2 = 91 per cent, 3 to 4 = 91 per cent, they were not significantly different. When followed serially in the same patient, during an episode of coma (Table 1), there was no apparent correlation with level of consciousness.

A comparison of oxygen saturation and CO_2 tension (Fig. 5) revealed an inverse correlation ($r = 0.29$) which was significant at the 5 per cent level of confidence.

Blood Urea Nitrogen in Relation to Level of Consciousness, Liver Function and Death. When group means for blood urea nitrogen were compared (grades: 0 = 12.4 mg./100 ml., 1 to 2 = 16.8 mg./100 ml., 3 to 4 = 41.1 mg./100 ml.) (Fig. 6), significantly higher values were observed in the most cerebrally obtunded group, $p = <0.01$.

Serial determinations were performed in ten patients who were observed during one or more episodes of coma. (Table 1.) Five patients had massive gastrointestinal hemorrhage; in two (T. L., W. H.) prominent azotemia was associated with marked liver dysfunction, oliguria and death (patient P. C., not shown, also exhibited these features). Two hemorrhaging patients (F. C., V. C.), whose initial tests on the liver were minimally to moderately deranged, showed transient elevations in urea nitrogen coincident with surgery; both survived. The fifth patient with hemorrhage (J. B., Table 1), who also lived, had no elevation in urea nitrogen; liver function was only minimally to moderately impaired and the urine volume was excellent. Pronounced azotemia was observed in three

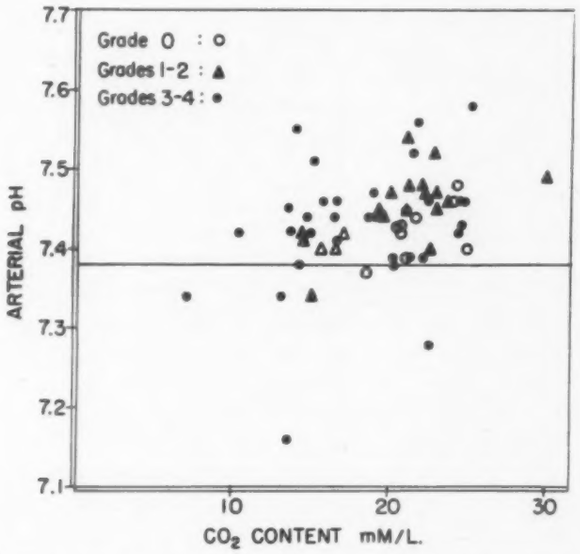

FIG. 4. Arterial pH in relation to carbon dioxide content in patients with liver disease at three levels of consciousness (sixty-one observations in twenty-three patients). Horizontal line represents mean pH for normal control subjects.

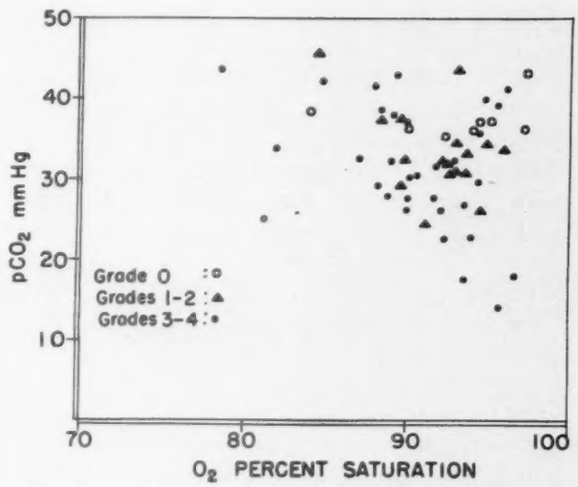

FIG. 5. Arterial oxygen saturation in relation to carbon dioxide tension in patients with liver disease at three levels of consciousness (fifty-seven observations in twenty-three patients).

patients (W. B., J. W., H. R.), and a modest elevation in a fourth (W. E.) who exhibited marked liver dysfunction and no evidence of gastrointestinal hemorrhage. Single observations in three other patients (G. S., Table 1, and W. M., G. W. not shown), in whom these features were noted, revealed prominent elevations in urea nitrogen. All but two (H. R., W. F.) of these seven patients were oliguric and died within one week of their last sampling. Only one patient (L. P.) died in hepatic coma of liver failure without azotemia.

COMMENTS

It is apparent from a consideration of the presented data and the observations of others that patients with liver disease, with all grades of consciousness, may exhibit multiple biochemical and blood gas defects, suggesting a derangement in several organ systems [*2,6,7,11,30,31*]. Nevertheless, when such patients were grouped in relation to their degree of coma, a progressive increase in arterial ammonia concentration with increasingly disordered mental status was observed. Overlap of arterial ammonia values between groups of patients at all levels of consciousness was present [*32–34*]. Overlap was less evident when individual patients were studied serially during a single episode of coma. The observations made over a one-year period in patient W. F. (Table 1) were of particular interest in this regard.

The blood ammonia may be contributed to by several sources [*32*]. The gastrointestinal tract as a major source of blood ammonia is variably affected by the nitrogenous load [*2,35*], oral broad-spectrum antibiotics [*21*], the functional

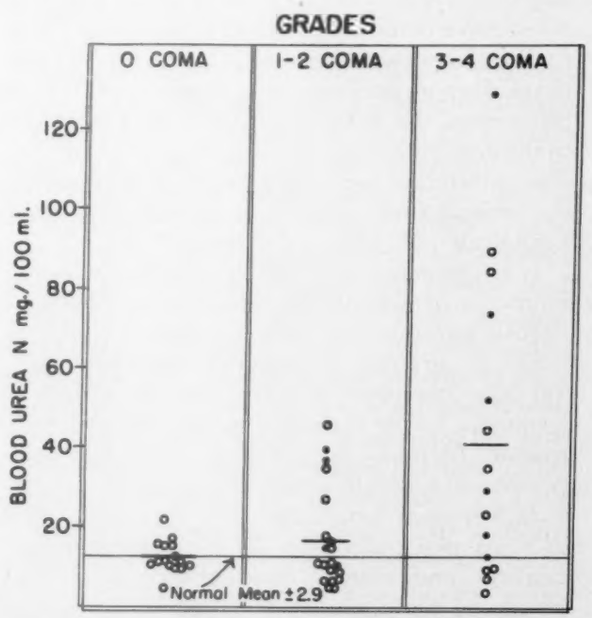

FIG. 6. Blood urea nitrogen concentrations in patients with liver disease at three levels of consciousness (fifty observations in thirty-three patients, using the highest concentration obtained from any one subject during an episode of coma). Closed circles represent samples obtained during gastrointestinal hemorrhage.

capacity of the liver and the presence and extent of portocaval communications [*36*]. Patients without liver disease do not exhibit significant diurnal fluctuations of blood ammonia concentration when fasting [*36*] or with normal dietary intake [*22*]. In the present study, alert patients with liver disease had significantly greater arterial ammonia concentrations than normal control subjects, confirming the observations of Webster and Gabuzda [*33*]. However, most of these patients, graded 0, were eating 80 to 90 gm. of protein daily and were not receiving oral broad-spectrum antibiotics. Alert patients who were being treated with protein withdrawal and antibiotics had arterial ammonia concentrations which were similar to normal subjects [*32*]. The present study yields no information regarding the relative importance of the liver and portocaval communications as routes by which gastrointestinal ammonia may gain entrance into the systemic circulation.

The arterial ammonia concentration of all patients with hepatic coma, grades 1 to 4, was greater than 1 standard deviation from the mean value for control subjects, in spite of protein withdrawal and oral broad-spectrum antibiotics. Indeed, despite this regimen, the highest arterial ammonia concentrations were observed in patients who were in grade 4 coma and had progressive deterioration of liver function. These findings in the absence of gastrointestinal hemorrhage support previous observations that sources other than the gastrointestinal tract contribute to the arterial ammonia concentration [*13,21,32*]. The extremities were not a source of ammonia in the present study. Additional sources may have been brain [*32*], kidney and liver [*21,36*].

It is apparent that the peripheral uptake of ammonia in patients with disordered consciousness is variable and may be great enough to lower the arterial ammonia concentration [*37*]. We have previously reported that the uptake of ammonia by the extremity in patients with liver disease becomes less obvious with increased peripheral blood flow [*38*]. The positive correlation between peripheral A-V ammonia and A-V oxygen differences in the present report confirm and extend these observations. The decreased peripheral A-V oxygen difference in patients with liver disease has been previously noted by others [*7,10*]. In the present study, the lowest A-V oxygen difference (0.05 volumes per cent) was observed in an alert patient with cirrhosis whose arterial ammonia concentration was 109 μg./100 ml. and whose cardiac index was 7.5 L./minute/M^2. The highest A-V oxygen difference (6.05 volumes per cent) was observed in a comatose, hemorrhaging patient with cirrhosis whose arterial ammonia concentration was 350 μg./100 ml. However, a consistent relationship between A-V oxygen difference and either arterial ammonia concentration or level of consciousness was not apparent. The data indicate that both the rate of peripheral blood flow and the arterial ammonia level are important in determining the A-V ammonia difference. Thus the positive correlation between peripheral A-V ammonia difference and arterial ammonia concentration was confirmed [*37,38*]. The group data suggest that significant peripheral ammonia uptake occurred in patients with liver disease who had moderate elevations of arterial ammonia concentration, grades 1 and 2, and that a relatively smaller increment was extracted in the periphery with further arterial ammonia elevation, grades 3 to 4. A similar sequence of peripheral ammonia uptake in relation to arterial ammonia concentration has been reported in normal subjects given intravenous ammonium salts [*24*]. In addition, the present data confirm the closer correlation of arterial ammonia concentrations than venous ammonia values with the presence and extent of hepatic coma [*32,33,37*].

Previous reports have commented upon the respiratory alkalosis and marked hyperpnea exhibited by some patients in hepatic coma [*6,15*]. In the present study a respiratory alkalosis was found in patients with liver disease at all levels of consciousness. The patients graded 0 did not show clinical hyperpnea. Four of the nine observations in this group were obtained from patients who had recently recovered from coma. However, those patients without previous alterations in consciousness showed similar values for pH and pCO_2. The initial observations made on patient W. F. (Table 1), who was eating 90 gm. of protein per day, were particularly significant. In contrast, deep blowing respirations were increasingly apparent with progressive coma and invariably present in grade 4 coma. This was reflected by the further reductions in pCO_2 which were observed in these patients. Although arterial pH appeared to be similar in all patients with liver disease when grouped according to state of consciousness, higher values were seen with progressive coma in some persons studied serially. However, four of five patients studied

within twenty-four hours of death showed values which were considerably lower than those previously obtained; two were frankly acidotic. The reduction in CO_2 tension was consistent with the presence of a respiratory alkalosis in patients with liver disease at all levels of consciousness, but the alteration was most pronounced in patients with cerebral dysfunction [*15*].

Studies in dogs and mice have suggested that ammonia toxicity may be potentiated by a coexistent alkalosis [*39,40*]. Intravenous infusion of ammonium salts (chloride and lactate) in normal human subjects was not accompanied by neurological or electroencephalographic alterations during the one-hour infusion period, despite arterial ammonia concentrations of 400 to 870 μg./100 ml. and the concomitant appearance of a modest alkalosis in three patients; maximum of 7.44 [*24*]. However, neurological and electroencephalographic changes consistent with those seen in hepatic coma have been reported in a patient with metabolic alkalosis (pH 7.56) and normal liver function eighteen hours after a continuous intravenous infusion of ammonium chloride [*41*]. Vanamee and associates have stated that their patients with liver disease who had coexistent elevations in ammonia concentration and reductions in pCO_2 were consistently in coma or on the verge of coma [*15*]. The effects of alkalosis on the NH_3-NH_4^+ buffer system and blood-brain barrier have been recently discussed [*39,40*]. From these observations it would appear likely that the disordered consciousness exhibited by patients graded 1 to 4 in this study was in part due to their alkalosis. Nevertheless, overlap of arterial ammonia concentrations between these groups did not appear to be explained by a consideration of ammonia concentration and pH *per se*. Additional observations on patients with liver disease at all levels of consciousness appear to be necessary.

It has been suggested that ammonia is a respiratory stimulant [*15*]. This statement has gained support from the observed hyperventilation and respiratory alkalosis in unanesthetized dogs and mice following the intravenous infusion of non-acidifying ammonium salts [*39,40*]. In addition, an increased respiratory rate and volume was evident in the records of human subjects given intravenous injections of 26 mEq. of ammonium chloride "as rapidly as possible," by Marossero and associates [*42*]. The mechanism of this effect remains unclear. Current theories include that of respiratory stimulation which may be secondary to cerebral anoxia or decreased oxygen utilization [*15,40*]. There has been no evidence that ammonia directly stimulates the respiratory center.

A reduced arterial oxygen saturation has been observed in some patients with liver disease in the absence of primary disease of the heart or lung [*7,8*]. A review of these data indicates a marked variation in the presence and degree of hypoxemia found in such patients [*7,10,31*]. This variability was apparent in our experience and was independent of the level of consciousness, although the alert group tended to have somewhat higher values than obtunded patients. A decrease in arterial oxygen saturation may be related to transient areas of pneumonitis, atelectasis or edema. Elevation of the diaphragm with ascites may also be a factor in producing moderate hypoxemia [*43*]. The finding of arterial oxygen unsaturation in the presence of lowered blood carbon dioxide tension suggests some type of process in which unsaturated blood is shunted by the well ventilated lung. Recently, pathological studies have demonstrated anastomoses between the portal circulation and the left side of the heart [*44*]. In some patients ventilation is impaired and hypoxemia and hypercapnea occur during coma.

The oliguria and azotemia seen in terminal patients with liver disease has been commented upon in several publications [*5,6,11*]. The present study was consistent with these observations in that both features were exhibited by eight of nine patients who were studied within a week before death. Although transient and at times progressive gastrointestinal hemorrhage and hypotension were observed in some patients, the presence of oliguria and an elevated blood urea nitrogen were at times independent of these complications. However, the nine patients who died had progressive bilirubinemia and pronounced reductions in prothrombin time unresponsive to vitamin K. In addition, hyponatremia was frequently observed. These findings in terminal patients with liver disease have been noted by others [*45*]. An elevation in urea nitrogen was not always a poor prognostic sign, as evidenced by the four patients with significant azotemia who survived; all maintained an excellent urinary output and bilirubinemia was marked only in one (H. R.). Although transient or prolonged arterial hypotension may be a

major factor in many patients [*46*], the mechanism of the azotemia exhibited by some patients remains unclear.

SUMMARY AND CONCLUSIONS

1. Biochemical and blood gas measurements were obtained in forty-nine patients with a variety of liver diseases. Group and serial determinations were compared in relation to level of consciousness.

2. A progressive increase in arterial ammonia concentration with increasing cerebral dysfunction was observed. Overlap of arterial ammonia values was present but was less evident when serial determinations were obtained from individual patients studied at different levels of consciousness.

3. The presence and extent of coma correlated more closely with arterial than peripheral venous ammonia concentrations. The extremity A-V ammonia difference was correlated with the height of the arterial level and the degree of peripheral A-V oxygen difference. It was concluded that peripheral A-V ammonia difference was controlled by two variables; (1) the concentration of ammonia in the arterial blood and (2) the quantity of blood flowing through the extremity.

4. Patients with cerebral dysfunction exhibited a significantly greater respiratory alkalosis than alert patients. It appeared likely that the alkalosis contributed to the disordered consciousness. However, its concomitance did not *per se* account for the occasional overlap between arterial ammonia concentrations and level of consciousness noted in individual patients.

5. Marked variation of arterial oxygen saturation was observed in this study. This variability was explained by the multiple processes which affect pulmonary circulation and ventilation in such patients.

6. None of the findings noted were necessarily of prognostic significance. However, the presence of azotemia with associated oliguria was seen only in patients who died.

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Hepatic Involvement in Extrapulmonary Tuberculosis*

Histologic and Functional Characteristics

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ALMOST a century ago Virchow and his contemporaries described miliary tubercles of the liver in tuberculous patients in whom autopsies were performed [*1*]. Until recently the liver seldom has been considered to be a site of tuberculous involvement except in cases of acute miliary tuberculosis or as a result of terminal hematogenous dissemination in far advanced pulmonary tuberculosis [*2–4*].

Hepatic involvement in the more chronic forms of miliary tuberculosis has only recently received attention [*1*]. Hematogenous seeding of tubercle bacilli, most frequently originating from the lung, is the cause of most varieties of acute or chronic extrapulmonary tuberculosis [*5,6*]. It is, therefore, not surprising that tuberculous granulomas are found in needle biopsy specimens of the liver in both acute and chronic disseminated tuberculosis [*1,7,8*].

Previous studies of hepatic function tests and their relationship to histologic abnormalities in patients with tuberculosis have yielded no consistent patterns [*9–14*]. In this investigation, liver function tests were performed in fifty patients with extrapulmonary tuberculosis. Abnormalities of hepatic function and of plasma proteins were demonstrated in almost all patients. A high incidence of granulomas and other significant histologic changes were observed in the liver biopsy specimens of thirty patients.

TABLE 1
CLINICAL TYPES OF EXTRAPULMONARY TUBERCULOSIS

Type of Extrapulmonary Tuberculosis	Patients (no.)
Peritonitis	13
Miliary	6
Meningitis	3
Lymphadenitis with pulmonary lesion	5
Lymphadenitis without pulmonary lesion	10
Lymphadenitis with pleural effusion	2
Lymphadenitis with fistula-in-ano	1
Chest wall abscess	1
Abdominal wall abscess	1
Psoas abscess, adrenal insufficiency	1
Osteomyelitis, lumbar vertebrae, without pulmonary lesion	1
Osteomyelitis, wrist, with pulmonary infiltration	1
Epididymitis with orchitis	2
Hepatosplenomegaly, with pulmonary infiltration	2
Cerebral tuberculoma	1
Total	50

MATERIAL AND METHODS

The clinical types of extrapulmonary tuberculosis in the fifty patients studied are shown in Table 1. The diagnosis of tuberculosis was established by histological and/or bacteriological studies. Patients with pleural or pericardial effusion, but no other demonstrable extrapulmonary involvement were excluded from this study.

One or more liver function tests were performed in all patients. In thirty patients liver biopsy specimens were obtained. The tests of hepatic function included the bromsulfalein (BSP) excretion according to the modification of Mateer (using 5 mg./kg. and determining the degree of dye retention in forty-five minutes) [*15*], the thymol turbidity by the procedure of Maclagen [*16*] as modified by Shank and Hoagland [*17*], the cephalin flocculation by the method of Hanger, as modified by Neefe and Reinhold [*18*], the alkaline phosphatase by the Bodansky method

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TABLE II

ANALYSIS OF THE MICROSCOPIC CHANGES OF THE LIVER IN EXTRAPULMONARY TUBERCULOSIS

Patient	Non-Specific Inflammatory Infiltration	Focal Kupffer Cell Hyperplasia	Granulomas	Caseous Necrosis	Parenchymal Cellular Abnormality	Fat	Fibrosis
			Group I				
J. T. H.	+	0	0	0	0	0	+
			Group IIA				
P. L. V.	+	+	0	0	+	+	0
F. J. O.*	+	+	+	0	+	+	0
A. W.	+	+	+†	0	+	+	0
E. S. B.	+	+	+†	0	+	0	+
R. O. C.	+	+	+†	0	+	0	+
J. D.	+	+	+	+	+	0	+
J. A.	+	0	0	0	+	+	+
A. A.	+	+	0	0	+	+	0
W. A. P.	+	+	0	0	+	0	+
L. P. V.	+	+	+	+	+	+	+
			Group IIB				
M. S.	+	+	+	0	+	0	0
O. A. M.	+	0	0	0	+	0	0
O. A. M.*	+	+	+	+	+	0	+
C. B. F.	+	+	+	0	+	0	0
C. B. F.*	+	+	+	+	+	0	+
L. F.	+	+	+	0	+	0	+
W. R. H.	+	+	+†	0	+	0	+
T. R.	+	+	+	0	+	0	+
J. A. G.	+	+	+	+	+	0	0
J. A. G.*	+	+	+	0	0	0	+
F. R.	+	0	+†	0	+	0	+
			Group III				
D. P. F.	+	+	0	0	+	0	+
G. D.	+	+	+†	0	+	0	0
L. A.	+	+	+	0	+	0	0
C. A.	+	+	+	0	+	+	0
			Group IV				
G. E. H.	+	+	+†	0	+	+	+
C. G.	+	+	+	+	+	0	+
D. T. L.	+	+	+	0	+	0	+
R. J.	+	+	+	0	+	0	0
A. T. H.	+	+	+	+	+	0	+
S. G.	+	+	+	+	+	+	+
L. T.	+	+	+	+	+	0	0

* Indicates surgical biopsy.

† Indicates microgranuloma.

TABLE III

RESULTS OF LIVER FUNCTION TESTS

Patient	Diagnosis	Serum		Cephalin Flocculation	Thymol Turbidity	Serum Bilirubin (mg. %)	Bromsulfalein (% retention)	Serum Alkaline Phosphatase (Bodansky units)
		Albumin (gm. %)	Globulin (gm. %)					
	Group I: Normal Liver Function Tests							
R. K. W.	Axillary lymphadenitis	3.0	3.0	0	2.4	0.7	2.5	2.3
G. A. N.	Chest wall abscess	3.3	4.0	0	2.6	0.8	0	3.0
S. W.	Cervical lymphadenitis, pulmonary infiltration	3.7	3.7	...	4.2	0.2	0.5	3.5
R. J. C.	Psoas abscess, adrenal insufficiency	3.3	2.8	0	1.2	0.7	1.5	4.4
E. W.	Cervical lymphadenitis	3.5	3.6	0	2.6	0.4	0	4.1
J. T. H.	Peritonitis	4.8	2.5	0	1.6	1.1	5.5	2.0
O. M.	Abscess of abdominal wall	4.4	2.4	0	0.8	0.2	3.5	4.1
R. P.	Meningitis	2.8	3.3	...	....	...	3.5	....
	Group II: Predominant Abnormality of Bromsulfalein and Serum Alkaline Phosphatase							
	(A) Bromsulfalein Only							
P. L. V.	Cervical lymphadenitis, pulmonary infiltration	3.6	3.5	1+	3.4	...	14.0	....
F. J. O.	Peritonitis	2.5	2.5	N	N	1.7	9.3	N
J. P.	Peritonitis	2.7	4.2	2+	....	0.9	8.5	....
R. W.	Peritonitis	3.5	5.3	1+	3.8	0.8	9.3	2.8
A. W.	Epididymitis, orchitis	3.9	2.8	0	2.4	0.8	7.5	3.3
C. H.	Meningitis	4.5	2.8	0	1.6	0.8	6.5	3.1
H. H. B.	Meningitis	4.8	2.5	0	0.4	0.4	6.5	3.5
E. S. B.	Cervical lymphadenitis	4.5	2.9	0	2.2	0.3	16.0	3.2
J. D. D.	Osteomyelitis, lumbar vertebrae	4.1	3.3	0	2.0	0.7	6.5	2.8
R. O. C.	Cervical lymphadenitis	4.2	2.7	0	2.4	0.5	7.5	1.6
J. D.	Miliary	...	...	1+	3.1	...	9.0	....
G. G.	Peritonitis	...	...	1+	3.1	...	6.0	....
J. A.	Cervical lymphadenitis	4.7	3.1	...	2.6	0.5	19.5	1.4
A. A.	Peritonitis	3.3	3.8	0	3.6	1.2	6.5	2.0
W. A. P.	Cervical lymphadenitis	3.7	3.7	1+	2.4	0.3	7.5	1.4
L. P. V.	Cervical chest wall node	3.9	3.4	0	2.6	0.9	7.3	2.0
	(B) Bromsulfalein and Serum Alkaline Phosphatase							
H. W.	Cervical lymphadenitis	3.8	4.2	0	1.2	0.9	6.5	16.9
M. S.	Cervical node abscess	4.5	3.8	0	3.2	0.7	15.0	9.6
O. A. M.	Miliary	4.2	5.5	1+	6.2	...	15.0	23.7
C. B. F.	Miliary	3.6	3.3	2+	2.2	2.6	15.0	27.2
L. F.	Epididymitis, orchitis	3.3	3.6	3+	5.3	0.9	21.5	56.7
W. R. H.	Cerebral tuberculoma	3.9	3.7	0	4.7	0.9	33.	6.4
T. R.	Lymphadenitis	3.9	3.2	0	2.4	0.7	5.5	6.0
J. A. G.	Peritonitis	4.3	3.2	0	4.5	1.3	13.5	5.2
F. R.	Cervical lymphadenitis, miliary pulmonary lesions	4.3	2.6	0	3.0	3.7	12.	5.3
	Group III: Predominant Abnormality of Flocculation Tests and Hyperglobulinemia							
D. P. F.	Hepatosplenomegaly, pulmonary infiltration	3.2	5.6	4+	16.0	1.2	7.5	6.6
A. B.	Peritonitis	1.7	4.3	4+	32.7	0.8	6.4	0.8
G. D.	Peritonitis, pleurisy	4.8	3.8	4+	13.0	1.2	6.5	3.4
L. N.	Cervical lymphadenitis, pulmonary infiltration, pleurisy	3.6	5.6	4+	4.2	1.7	6.5	2.9
R. L. B.	Osteomyelitis of wrist, pulmonary infiltration	3.7	5.0	...	5.2	0.7	....	3.2
L. A.	Axillary lymphadenitis, pleural effusion	4.5	3.5	0	7.3	0.5	5.0	....
C. A.	Peritonitis	2.8	3.7	4+	8.7	2.5	2.5	4.1
	Group IV: Combinations of Groups II and III							
G. E. H.	Cervical lymphadenitis, pleurisy	3.1	4.8	4+	11.0	0.5	14.0	6.6
C. G.	Miliary	2.1	5.3	4+	16.6	2.3	19.5	38.6
J. T.	Peritonitis	2.8	4.1	3+	5.7	0.7	21.0	7.1
D. T. L.	General lymphadenitis	4.1	3.7	0	6.2	0.3	13.0	6.0
R. J.	Cervical lymphadenitis, pulmonary infiltration	3.6	4.2	3+	6.2	0.4	8.5	4.0
A. T. H.	Miliary	3.9	4.7	4+	2.8	0.9	26.	4.6
S. G.	Hepatomegaly, prominent pulmonary infiltration	2.8	4.0	4+	1.6	0.5	12.	16.9
J. S.	Peritonitis	3.9	4.1	3+	3.8	0.7	9.3	4.2
L. T.	Miliary	3.6	3.7	4+	5.7	1.2	19.5	8.1
W. Z.	Miliary	2.9	2.9	4+	6.2	5.0	58.7	17.7

TABLE IV
INCIDENCE OF ABNORMALITY OF LIVER FUNCTION IN EXTRAPULMONARY TUBERCULOSIS

Tests	Patients Studied (no.)	Abnormal (no.)	Abnormal (%)
Bromsulfalein retention	49	42	85.7
Cephalin flocculation	46	15	32.5
Thymol turbidity	48	19	39.6
Serum bilirubin	45	12	26.7
Alkaline phosphatase	44	18	40.9
Serum albumin	48	26	54.2
Serum globulin	48	36	75.0

[*19*], and the serum bilirubin by the Malloy and Evelyn method [*20*]. The level of the total serum proteins and the albumin and globulin fractions were determined by the Weichselbaum modification [*21*] of the Kingsley biuret method [*22*].

Biopsy of the liver was performed with the Vim-Silverman needle through the intercostal approach in twenty-seven patients. In three patients, biopsy specimens were obtained at the time of abdominal laparotomy. (Table II.) The tissue sections were prepared in the usual manner and stained with hematoxylin-eosin. Approximately fifteen sections were made from each specimen. In nine instances Ziehl-Neelsen stained sections for acid-fast bacilli were also prepared.

RESULTS

The results of the liver function tests were divided into four groups. (Table III.) Eight patients (group I) had essentially normal hepatic function and showed only serum protein abnormalities. Twenty-five patients (group II) had abnormal bromsulfalein excretion with or without alkaline phosphatase elevations and little or no abnormality in the results of other tests. Seven patients (group III) were characterized by predominant abnormality of the flocculation tests and hyperglobulinemia. The patients in group IV represented the combined abnormalities of groups II and III. Abnormal serum protein levels or abnormal results in liver function tests were seen in all patients, and hepatic dysfunction was noted in forty-two of the fifty patients.

An analysis of the individual tests (Table IV) showed that impairment of bromsulfalein excretion was the most frequent abnormality (85.7 per cent). The degree of dye retention was usually mild, with an average of 13 per cent. Only three patients had more than 25 per cent retention.

TABLE V
SUMMARY OF MICROSCOPIC CHANGES OF THE LIVER IN EXTRAPULMONARY TUBERCULOSIS
(30 PATIENTS)

Changes	Abnormal (no.)	Abnormal (%)
Non-specific inflammatory infiltration	30	100
Focal Kupffer cell hyperplasia	27	90
Granulomas	24	80
Fibrosis	20	67
Fatty metamorphosis	9	30

Elevated serum alkaline phosphatase levels were observed in eighteen (40.9 per cent) of the patients studied. Seven patients had levels over 15 Bodansky units and one patient had a striking level of 56 units.

Abnormal results in flocculation tests were observed in one-third of the patients studied. Thymol turbidity levels were as high as 32 units.

Hyperbilirubinemia was mild and relatively infrequent, occurring in one-fourth of the patients. Only three patients had clinical jaundice with bilirubin elevations to levels of 2.6, 3.7 and 5.0 mg. per cent, respectively.

Abnormality of the plasma proteins was observed in 83 per cent of the patients studied. Half of these patients showed mild to moderate degrees of hypoalbuminemia. Three-fourths of the group had elevated serum globulin levels, sixteen patients showing values of 4.0 gm. per cent or more, and six patients with levels over 5.0 gm. per cent. Electrophoretic analyses in ten patients revealed the globulin elevation to be primarily of the gamma fraction, although in several patients significant elevation of the alpha globulin was also seen.

The histologic abnormalities observed in the liver biopsy specimens of thirty patients with tuberculosis are presented in Table II and summarized in Table V. Abnormalities were noted in all specimens.

Granulomas were demonstrated in twenty-four (80 per cent) of the biopsy specimens. These ranged from large, conglomerate tubercles with central caseation and Langhans' giant cells (Fig. 1), to small granulomas without caseation or giant cells. (Fig. 2.) Very small granulomas, composed only of epithelioid cells, were designated as "microgranulomas." (Figs. 3 and 4.) These lesions appeared to be similar to the

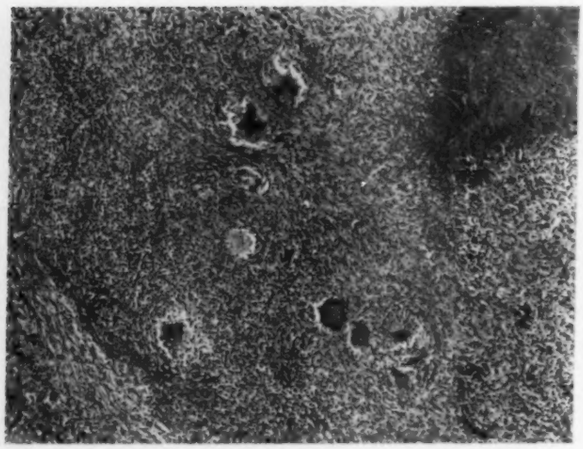

FIG. 1. Large conglomerate tubercles of the liver with central caseation and Langhan's giant cells.

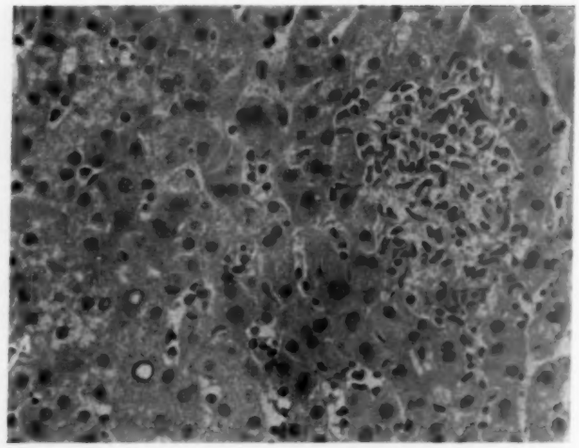

FIG. 2. Small epithelioid tubercle of the liver without caseation or giant cells. Note hydropic nuclei.

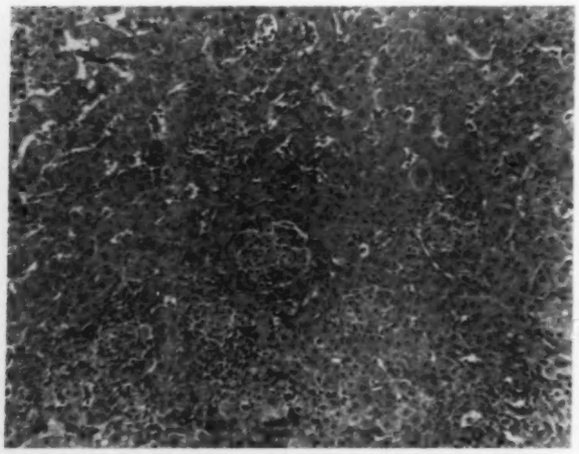

FIG. 3. Microgranulomas of the liver.

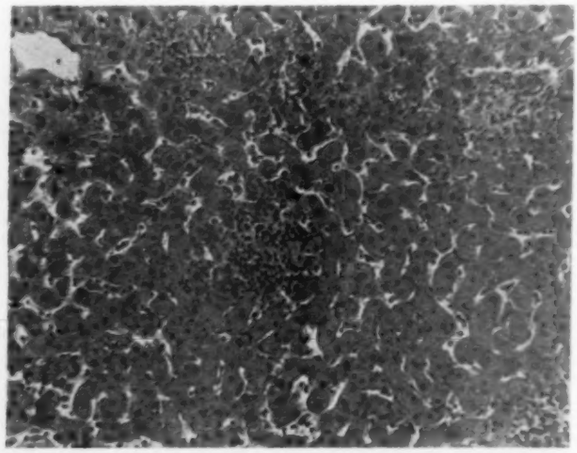

FIG. 4. Microgranuloma of the liver. Note diffuse sinusoidal inflammatory reaction.

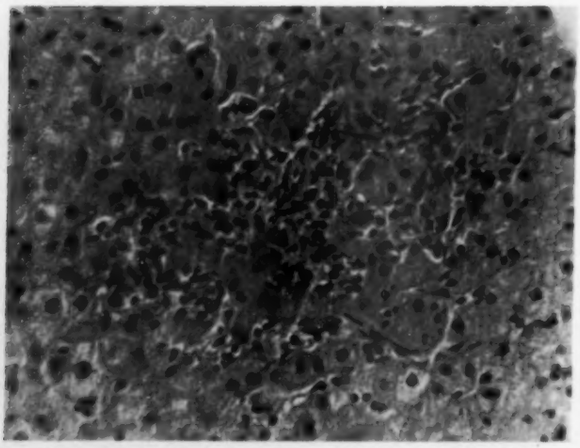

FIG. 5. Focal hyperplasia of Kupffer cells (retothelial or histiocytic nodule).

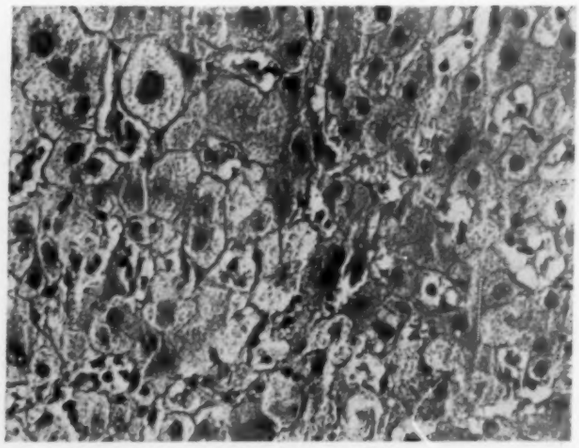

FIG. 6. Abnormalities of hepatic parenchymal cells.

subtubercles (types III and IV) of Haex and Van Beek [*1*]. The granulomas in sixteen of these twenty-four patients were easily recognizable while in eight patients only microgranulomas were seen.

Even more common (90 per cent) was the presence of stellate focal hyperplasia of Kupffer cells (retothelial or histiocytic nodules) [*11,23,24*]. These foci of proliferation (Fig. 5) were seen in distended sinusoids, associated with lymphocytic infiltration and often accompanied by necrosis of adjacent hepatic cells.

Focal or diffuse sinusoidal infiltration, predominantly lymphocytic, was observed in all patients studied. (Figs. 2 to 4.) The diffuse inflammatory reaction was prominent in some patients and the dense cellular infiltration in the portal areas of other patients was striking.

Abnormalities of the hepatic parenchymal cells (Fig. 6) were frequent but predominantly focal rather than diffuse. In degree they ranged from smudging of cell outlines and variability of cellular and nuclear size to frank focal necrosis. In three patients "free hyaline bodies" were seen. (Fig. 7.) Hydropic degeneration of nuclei was noted (Fig. 2) in seven patients.

Fatty metamorphosis of the liver was observed in nine (30 per cent) of our patients. In only one patient was it of a marked degree.

Fibrosis was noted in twenty (67 per cent) of the thirty patients. For the most part it was minimal and located in the portal and periportal areas. Architectural distortion to a degree compatible with cirrhosis was not observed in these biopsy specimens.

Ziehl-Neelsen stains were made in nine of the liver biopsy specimens. Acid-fast bacilli were observed in two patients with caseous granulomas.

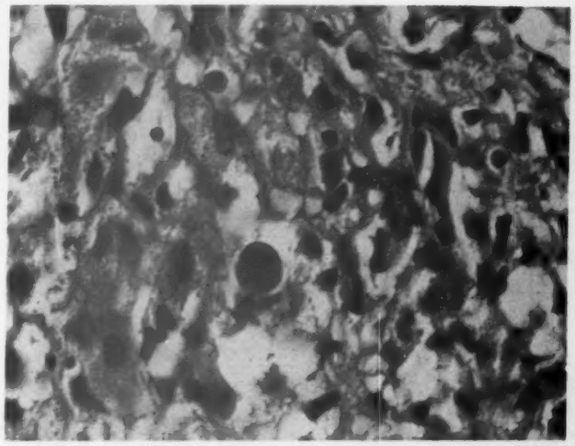

FIG. 7. Free hyaline body associated with necrosis.

COMMENTS

The high incidence of abnormal hepatic function tests in our patients is in agreement with other reports of patients with pulmonary and extrapulmonary tuberculosis. Some of these results are summarized in Table VI.

Impairment of bromsulfalein excretion was observed in 85 per cent of the patients in the present study. Klatskin and Yesner [*8*] also noticed this abnormality in three of four patients with acute disseminated tuberculosis. Other studies [*9,10,14,26–29*] of patients with pulmonary as well as extrapulmonary tuberculosis have revealed a varying incidence of bromsulfalein abnormality. Kruger and Gerber [*25*] have related the incidence of impaired bromsulfalein excretion to the severity of pulmonary tuberculosis.

Elevated values for serum alkaline phosphatase were observed in 41 per cent of our patients. Hurst et al. [*28*] and Klatskin and Yesner [*8*] also have reported a high incidence of this abnormality in patients with far-advanced pulmonary and disseminated tuberculosis, respectively. Other workers have reported a lower incidence of elevated alkaline phosphatase values. (Table VI.) Many of the patients studied by these authors had been or were under treatment. That levels of alkaline phosphatase may be markedly increased is illustrated by the patients of Klatskin and Yesner (40.4 Bodansky units) [*8*], Leonard, Katz, and McCormick (42 Bodansky units) [*30*], Ross, Iber and Harvey (98.5 Bodansky units) [*31*] and by one of our patients (56.7 Bodansky units). Each of these patients had extensive granulomatous infiltration of the liver. These observations are consistent with the views expressed by Ross et al. [*31*], that "infiltrative" lesions within the liver may produce extremely high levels of serum alkaline phosphatase.

The incidence of abnormal results of the thymol turbidity (39.6 per cent) and cephalin flocculation (32.5 per cent) tests observed in this study are consistent with the reports of Katz et al. [*27*], Galen et al. [*29*] and Seife et al. [*14*]. An even higher incidence has been reported by Ban [*10*] and Klatskin and Yesner [*8*].

The observation of mild and relatively infrequent hyperbilirubinemia noted in our patients is in agreement with most authors. Marked jaundice in patients with tuberculosis has been

TABLE VI
HEPATIC DYSFUNCTION IN TUBERCULOSIS

Author	Patients Studied (no.)	Bromsulfalein Retention % Abnormal	Cephalin Flocculation % Abnormal	Thymol Turbidity % Abnormal	Serum Alkaline Phosphatase % Abnormal	Type of Tuberculosis (comments)
Korn et al.	50	85.7	32.5	38.6	40.9	Extrapulmonary
Klatskin and Yesner [8]	4	75	100	75	67	Disseminated
Ban [10]	60	67	51.7	75	0	Pulmonary
Before treatment	35	71	.....	80	0	.....
After treatment	25	60	.....	68	0	.....
Kruger and Gerber [25]						
Far advanced	13	54	.....	....	....	Pulmonary (pre-chemotherapy era)
Moderately advanced	12	8.3	.....	....	....	
Minimal with amyloidosis	20	90	.....	....	....	.....
Levinson and Siegel [26]	28	46	.....	....	....	Far advanced pulmonary and extrapulmonary
Katz et al. [27]	25	24	16	28	0	Pulmonary (after chemotherapy and during pneumoperitoneum)
Hurst et al. [28]	17	23	.....	....	55	Far advanced pulmonary
Galen et al. [29]	53	19	30	....	....	Pulmonary
Seife et al. [14]	70	15	.....	30	15	Pulmonary (some during treatment)
Arora et al. [9]	102	....	54	23	....	Pulmonary and extrapulmonary

sufficiently unusual to have been the subject of separate case reports [32,33].

Hyperglobulinemia, demonstrated in 75 per cent of our patients, has been a frequent observation in pulmonary tuberculosis. Seife and associates [14] found a similar incidence (78 per cent) in their group of patients with active chronic pulmonary tuberculosis. Gohr and Klement [34] reported that all thirty-three of their patients had elevated globulin levels, predominantly involving the alpha and gamma fractions. Siebert and Nelson [35] studied electrophoretic patterns of the serum proteins of patients with tuberculosis. They commented upon the consistent elevations of gamma globulin and the occasional increase in alpha globulin. This is in accord with our results and other similar studies in the past few years [36].

Analysis of the results of hepatic dysfunction tests led to the formulation of the four categories shown in Table III. The pattern of hepatic dysfunction in group II is characteristic of patients with "space-occupying" lesions of the liver [37]. The latter include metastatic carcinoma [38], amebic abscess, amyloidosis and granulomatous infiltrative lesions [31]. Bromsulfalein retention, the most sensitive measure of hepatic dysfunction in clinical use, is evident in these patients as well as in those with febrile diseases [39].

The pattern of abnormality of the flocculation tests with little or no abnormality of the bromsulfalein or alkaline phosphatase values has been demonstrated in other hyperglobulinemic diseases, such as subsiding malaria [40], systemic lupus erythematosus [41], subacute bacterial endocarditis [42], sarcoidosis [43], and lymphogranuloma venereum [44].

The generalized hepatic dysfunction seen in the ten patients in group IV resembles that of patients with intrinsic hepatic disease, such as hepatitis.

The high incidence of granulomas in the present study (80 per cent) is consistent with the report of Klatskin and Yesner [8], Haex and Van Beek [1] and Mather et al. [4] concerning patients with extrapulmonary tuberculosis. (Table

TABLE VII
INCIDENCE OF HEPATIC GRANULOMAS BY BIOPSY

Author	Patients Studied (no.)	Granulomas (%)	Type of Tuberculosis
Klatskin and Yesner [8]	4	100	Miliary
Haex and Van Beek [1]	189	93	Pulmonary and extrapulmonary
Korn et al.	30	80	Extrapulmonary
Mather et al. [4]	22	68	Miliary and meningeal
Buckingham et al. [11]	22	40	Extrapulmonary
von Oldershausen et al. [45]	93	25.3	Miliary
Klatskin and Yesner [8]	13	23	Pulmonary and extrapulmonary
Ban [10]	59	20.3	Pulmonary
von Oldershausen et al. [45]	248	19.2	Pulmonary
Seife et al. [14]	70	13	Pulmonary
Arora et al. [9]	50	12	Pulmonary and extrapulmonary
Mather et al. [4]	34	3	Pulmonary
Van Buchem [46]	9	0	Pulmonary

TABLE VIII
INCIDENCE OF HEPATIC GRANULOMAS AT AUTOPSY

Author	Patients Studied (no.)	Granuloma (%)	Type of Tuberculosis
Torrey [47]	33	100	Miliary
Lorentz [48]	100	99	Pulmonary and extrapulmonary
Saphir [49]	100	80	Pulmonary
Ullom [50]	100	79	Pulmonary and extrapulmonary
Torrey [4]	89	50	Pulmonary
Stemmerman [51]	1500	32	Pulmonary and extrapulmonary

VII.) Studies of needle biopsies performed in patients with pulmonary tuberculosis have shown a lower incidence of hepatic granulomas [45,46]. This difference has been emphasized by Mather et al. [4] in a study of needle biopsies and by Torrey [47] in a study of autopsy material.

Nevertheless, there has been variation in the reported incidence of hepatic granulomas in patients with tuberculosis [48–51]. (Tables VII and VIII.) A number of factors may be responsible for the differences; these include the varying severity of the disease, the modifying effect of antituberculous chemotherapy and the differing criteria employed in defining granulomas. Haex and Van Beek [1] have considered their exhaustive search (over 100 sections per liver biopsy specimens) responsible for the high incidence of granulomas reported by them. Furthermore, their observations were based on a precise and novel classification of granulomas. They classified granulomas into five types, depending upon their size, location and characteristics. The smallest type, which they called "epithelioid cell sub-tubercle," is 0.05 mm. in diameter, is centrolobular in location and lacks giant cells and caseation. These lesions are similar, we believe, to those we have called microgranulomas. Unawareness of lesions of this size, which have not been emphasized by other workers, may explain the low incidence of granulomas in some reports.

Another histologic abnormality in our liver biopsy specimens was the presence of focal hyperplasia of Kupffer cells, called "retothelial nodules" by Hamperl [23] and Ringleb [24]. This term refers to localized areas of Kupffer cell hyperplasia within dilated sinusoids and radiation into adjacent sinusoids. Buckingham and his associates [11] have used the term "histiocytic nodules" in describing similar lesions. Popper and Schaffner [13] also have emphasized this focal Kupffer cell proliferation. Although such lesions may be forerunners of microgranulomas, they differ from the smallest granuloma in that they are composed of typical Kupffer cells, have a stellate rather than rounded configuration, and lack characteristic epithelioid cells.

A consistent feature of all biopsies was an inflammatory reaction, predominantly lymphocytic in nature. This reaction was characteristically diffuse and sinusoidal, but often was accompanied by focal infiltration associated with focal necrosis. This type of inflammatory reaction, observed in miscellaneous systemic diseases, has been called "non-specific reactive hepatitis" by Buckingham et al. [11] and Popper and Schaffner [13].

Abnormalities of the hepatic parenchymal cells were seen in almost all biopsy specimens in this study. In some of these the changes were slight and focal; in others there was evidence of necrobiosis and necrosis and even free hyaline bodies were seen. "Irregular hepatocellular damage" has been included by Popper and Schaffner [13] in their description of non-specific reaction of the liver to the presence of systemic diseases. We have seen no previous reference, however, to the occurrence of free hyaline bodies in patients with tuberculosis. This morphologic feature, considered [52] to be the equivalent of the Councilman body of yellow fever, is characteristic of viral hepatitis. Similar acidophilic bodies have been observed in patients with

TABLE IX
INCIDENCE OF FATTY METAMORPHOSIS

Author	Patients Studied (no.)	Fatty Metamorphosis (%)	Type of Tuberculosis
Parini [60]*	50	100	Pulmonary and intestinal
Jones and Peck [61]*	581	41.8	Pulmonary and extrapulmonary (intestinal, 53%)
Ban [10]†	59	37.3	Pulmonary
Ullom [50]*	100	35	Pulmonary and extrapulmonary
Saphir [49]*	100	34	Pulmonary
Arora et al. [9]†	50	28	Pulmonary and extrapulmonar y
Korn et al.†	30	30	Extrapulmonary
Seife et al. [14]†	70	20	Pulmonary

* Autopsy study.
† Biopsy study.

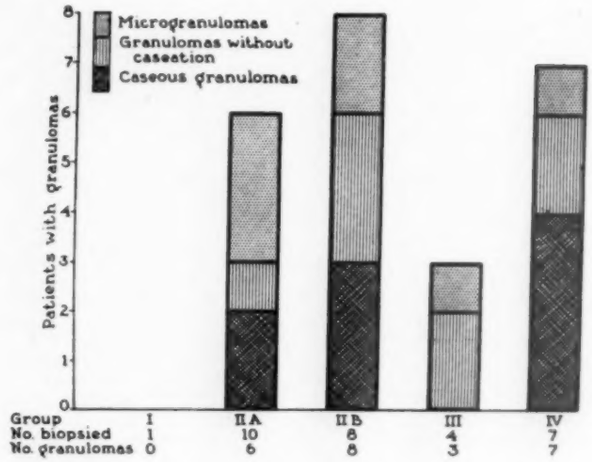

FIG. 8. The type of granulomas which were demonstrated in twenty-four of the thirty patients in whom biopsies were performed, correlated with patterns of liver function.

hepatic damage caused by toxic hepatitis, burns, tannic acid, congestive heart failure and obstructive jaundice [52]. We would add hematogenous tuberculosis to this group. The term "granulomatous hepatitis" might seem appropriate in those patients whose livers showed marked hepatocellular abnormalities as well as inflammatory and granulomatous infiltration.

Portal fibrosis was observed in 67 per cent of the biopsy specimens studied, but there was insufficient architectural distortion for a diagnosis of cirrhosis. This is consistent with the views of Eppinger [53], Karsner [54] and Klatskin [12] although a pathogenetic relationship between tuberculosis and cirrhosis has been discussed frequently in the past [2,55,56]. Other granulomatous diseases involving the liver have been reported to lead to cirrhosis. McCullough and Eisele [57] observed such changes in a patient who was seriously ill with brucellosis; serial biopsies of the liver showed early and acute granulomatous hepatitis and later a coarse nodular or postnecrotic cirrhosis. Spink and co-workers [58] also concluded that there is accumulating evidence that the granulomatous hepatitis of brucellosis can lead to cirrhosis. Similarly in sarcoidosis, sufficient fibrosis with architectural distortion of the liver to produce portal hypertension has been reported [6,59].

In the past, tuberculosis was considered a common cause of fatty metamorphosis of the liver [2,3]. This lesion was seen in nine (30 per cent) of our patients, but in four of these patients the amount of fat was minimal. These observations agree with recent reports based on needle biopsy studies. (Table IX.) Haex and Van Beek [1] in their monograph did not refer to the presence of fat in their liver biopsy specimens.

TABLE X
CORRELATION OF HEPATIC DYSFUNCTION WITH HISTOLOGIC ABNORMALITIES

Group	Patients Studied (no.)	Granulomas	Caseous Necrosis	Moderate Inflammatory Infiltration	Moderate Cellular Abnormality	Fat	Fibrosis
Group I (normal)	1	0	0	0	0	0	1
Group IIA (bromsulfalein only)	10	6	2	2	5	5	7
Group IIB (bromsulfalein and serum alkaline phosphatase predominantly)	8	8	3	5	4	1	6
Group III (flocculations predominantly)	4	3	0	2	1	1	1
Group IV (combined abnormalities)	7	7	4	4	5	2	5

Parini [*60*] and Jones and Peck [*61*] noted the frequency of fatty metamorphosis of the liver at autopsy in intestinal and far-advanced pulmonary tuberculosis. Jones and Peck, however, and other authors have commented on the role of malnutrition in the pathogenesis of fatty metamorphosis [*9,12,14,56*]. It is also our impression that fatty metamorphosis is coincidental and not directly related to the tuberculosis.

The pathogenesis of these hepatic lesions in tuberculous patients may be clarified by comparison with similar abnormalities observed in certain experimental animals. Different local tissue conditions, host resistance and organism virulence determine the extent of tuberculous involvement of a particular organ. Lurie [*62*] and Wessels [*63*], working with rabbits and rats, respectively, have shown that there is less apparent survival of tubercle bacilli in the liver than in the lung. Caseous granulomas were less frequent in the liver than in the lung. Hepatic granulomas with caseation, when present, contained demonstrable tubercle bacilli. Lurie also noted that the formation of small, epithelioid, non-caseous granulomas was associated with the disappearance of tubercle bacilli from the liver. In the present study, the most frequent lesion was the granuloma without caseation. It is of interest, however, that all six patients with acute or subacute miliary tuberculosis had larger caseous granulomas. These results are consistent with the view that an overwhelming tuberculous dissemination is more likely to be associated with hepatic caseous granulomas. It is reasonable to infer that less extensive dissemination results in non-caseous smaller granulomas.

An attempt has been made to correlate the patterns of hepatic dysfunction with the histological abnormalities of the liver in the thirty patients in whom biopsy specimens were obtained. (Table x, Fig. 8.) Granulomas were usually demonstrated when the alkaline phosphatase value was elevated (fourteen of fifteen patients). The converse, however, did not hold. In ten of the fifteen patients with normal alkaline phosphatase levels, granulomas were also demonstrated. Twenty-two of the twenty-seven patients with impaired bromsulfalein excretion had granulomas. Larger granulomas with caseation were more frequent in the patients with greater hepatic dysfunction (group IV). No other consistent correlation between histologic changes and hepatic dysfunction was evident.

SUMMARY

Hepatic function was studied in fifty patients with extrapulmonary tuberculosis. Abnormalities of one or more of the liver function and/or serum protein tests were found in all patients. The most frequent changes were impaired bromsulfalein excretion and hyperglobulinemia. Analysis of these results revealed several patterns of hepatic dysfunction which have been described previously with other diseases. These patterns are (1) the elevated serum alkaline phosphatase and bromsulfalein retention associated with space-occupying lesions (granulomas), (2) the abnormal flocculation tests and hyperglobulinemia associated with certain chronic infections and inflammatory diseases and (3) combinations of (1) and (2) which may simulate intrinsic hepatic disease except for the relatively lower levels of serum bilirubin.

Liver biopsy specimens were obtained in thirty of these patients. Granulomas were demonstrated in twenty-four or 80 per cent of the specimens. Fourteen of the fifteen patients with elevated alkaline phosphatase levels had hepatic granulomas. Ten of the fifteen patients without alkaline phosphatase elevations also had granulomas. Microgranulomas were described and were considered significant. Focal Kupffer cell hyperplasia and diffuse sinusoidal inflammatory reaction were sufficiently frequent to be characteristic of hepatic involvement in tuberculosis. Abnormality of the hepatic parenchymal cells was commonly observed and in three patients free hyaline bodies were present.

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Vitamin B_6 Deficiency and Oxalate Nephrocalcinosis in the Cat*

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KNOWLEDGE of the origin of the large quantities of oxalate found in human renal calculi and associated with oxalosis and other disease states is limited. The present study of vitamin B_6 deficiency in cats provides a new approach to this problem.

EXPERIMENTS

Three- to six-month old kittens of both sexes were used in these experiments. Before being admitted to the laboratory they were dusted with an insecticide, dewormed and vaccinated against feline distemper. The cats were housed individually in metal mesh cages, and food and water were maintained *ad libitum*. The diet fed contained, percentagewise, casein 32.1, sucrose 37.6, corn oil 12.5, hydrogenated fat 12.5, cod liver oil 1, salts IV [*1*] 4 and choline 0.3 per cent. Four milligrams of thiamine hydrochloride, 8 mg. riboflavin, 40 mg. niacin, 20 mg. calcium pantothenate, 1 mg. folic acid, 0.2 mg. biotin and 1 mg. of menadione were added to each kilo of diet.

The cats were divided into groups and fed pyridoxine hydrochloride supplements of 0, 1 or 2 mg. per kilo of diet. There was also available for study material from a large number of control cats from other experiments, in which the basic diet contained 4 mg. of vitamin B_6 per kilo.

Periodically during the experimental period determinations of hemoglobin, hematocrit, red blood cells, white blood cells, urinary oxalic acid [*2*] and inorganic sulfate [*3*] were made. The animals were sacrificed (intravenous Nembutal®) after three to six and a half months on the experimental diets. One cat (No. 164) died after two months. Complete autopsies were performed and the tissues were fixed in 10 per cent neutral formalin and stained with hematoxylin and eosin. Brain, spinal cord and selected peripheral nerve sections (brachial and sciatic) were stained with luxol fast blue. Additional stains included Sudan IV, Gomori's ferrocyanide, periodic acid-Schiff, Giemsa, von Kossa, cresyl violet, alizarin red S and alcian blue.

RESULTS

Acute vitamin B_6 deficiency, characterized by growth failure, emaciation, convulsions and anemia, was produced in two to two and a half months in all four of the cats given an unsupplemented diet. Only one of the four cats receiving 1 mg. of vitamin B_6 per kilo of diet developed convulsions; this was followed by weight loss. The severe anemia present in the cats on an unsupplemented diet (Table I) could be considerably modified by the addition of 1 mg. of vitamin B_6 per kilo of diet. Data on growth and blood levels in cats receiving 2 mg. vitamin B_6 per kilo of diet appeared to be within the normal range when compared with control cats in this experiment and cats fed similar purified control diets over the past five years.

The most striking chemical finding was the presence in the urine and kidneys of large quantities of oxalate, as determined by the spot tests described by Feigl [*4*]. Table I shows a marked inverse relationship between the dietary vitamin B_6 content and the level of urinary and kidney oxalate, as determined by the method of Powers and Levatin [*2*]. A urinary component that appeared to be quantitatively related to the

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TABLE I

EFFECTS OF FEEDING DIETS DEFICIENT IN VITAMIN B_6 TO CATS

Diets	Cat. No.	Hemoglobin (gm./%)	Hematocrit (vol. %)	Red Blood Cells ($\times 10^6$/cu. mm.)	White Blood Cells (/cu. mm.)	Urinary Oxalic Acid (mg./kilo/24 hr.)	Kidney Oxalic Acid (mg./gm.)	Urinary Inorganic S (mg./kilo/24 hr.)
0 B_6/kilo diet.....	161	4.4	11	5.17	7000	23.4	1.14	27
	162	2.9	11	1.83	23800	24.1	12.20	21
	163	7.8	18	6.00	12400	....	4.17	40
	164	5.7	13	3.55	8500	....	.....	...
1 mg. B_6/kilo diet.....	165	7.4	22	7.04	7640	7.3	0.05	51
	166	11.6	33	12.26	4600	6.4	0.24	54
	167	7.6	21	7.16	16360	6.9	0.10	45
	168	10.2	28	8.96	6440	6.2	0.70	41
2 mg. B_6/kilo diet.....	169	11.5	28	8.73	9800	2.0	0.10	74
	170	12.0	31	10.71	11920	4.1	0.13	65
	171	9.9	24	8.49	13240	4.2	0.12	103
	172	12.3	30	11.00	8120	4.4	0.12	61
4 mg. B_6/kilo diet.....	147	10.1	22	5.30	10600	2.6	.....	...
	153	10.4	24	9.99	9600	2.2	.....	...
	157	11.9	28	8.90	16800	2.3	.....	...
	158	13.8	33	11.35	22300	0.6	.....	...

vitamin B_6 content of the diet was inorganic sulfate, which was excreted in increasing amounts as the vitamin B_6 content of the diets was increased.

The most significant pathologic changes were seen in the kidneys of the vitamin B_6 deficient animals. Renal scarring was related to the kidney oxalate content. The lesions consisted of cortical scars with flat deep brown bases. These were usually irregular in shape, 2 to 4 mm. in width although occasionally narrow and linear. Generally, approximately 25 per cent of the cortical surface was involved in this process although in one animal, No. 162, surface involvement approached 75 per cent. These changes were seen in all four of the cats on an unsupplemented diet and in three of the four cats receiving 1 mg. of vitamin B_6 per kilo of diet. None of the cats receiving 2 mg. of vitamin B_6 supplement per kilo of diet showed renal lesions. No gross calculi were seen at any level of the urinary tract.

Microscopically the scars were sharply circumscribed and pyramidal, the base representing the depressed cortical surface and the apex extending into the inner cortex and in some instances well into the medulla. (Figs. 1 and 2.) The scars appeared to be due largely to atrophy and loss of cortical tubules, with marked condensation of parenchyma. Throughout the scarred areas there was a minimal degree of tubular dilatation, of patchy distribution. The epithelium of both the small atrophic and the dilated tubules was amphophilic, and that of the latter in particular was flattened. The epithelium was rarely hyperplastic. Mitoses were uncommon. The glomeruli on occasion showed a remarkable concentration due to tissue condensation but no intrinsic change. (Fig. 1.) With periodic acid-Schiff staining, thickening of glomerular capsules and tubular basement membranes was seen sporadically within the scars. Scattered arterioles showed a non-hyaline, hyperplastic thickening of their walls. (Fig. 5.)

Within the cortical scars and extending into the medulla were large numbers of birefringent crystals. Because of the alterations of the tubular epithelium, it was difficult to localize accurately the position of the crystals within the nephron. Even in instances where crystal deposition without scarring appeared to be recent, focal tubular epithelial atrophy was generally seen. Crystals were most prominent in the cortex and in the distal collecting tubules. (Fig. 2.) In the latter location crystals were occasionally associated with considerable tubular dilatation (Fig. 3) and, although the epithelium of such tubules was generally flattened and appeared atrophic, instances of focal hyperplasia and epithelialization of crystals were seen. (Fig. 4.) In several

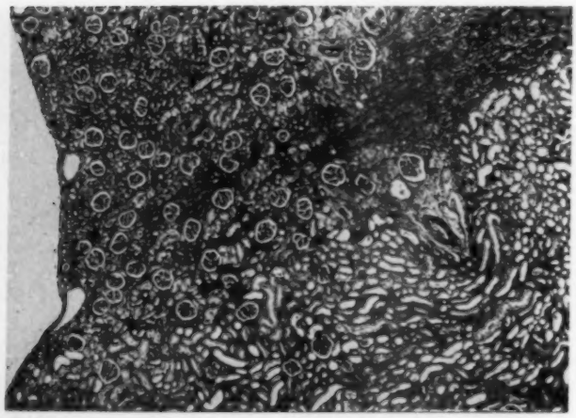

FIG. 1. Low power view of cortex showing boundary of normal cortex and discrete scar which extends into the medulla. There is marked loss of tubules with condensation of normal-appearing glomeruli. The residual tubules are small and atrophic. Tubular dilatation, inflammation and proliferation of fibrous tissue are absent. Hematoxylin and eosin stain, original magnification × 30.

cases small crystalline masses were present beneath the epithelium of the renal pelvis or ureter. (Fig. 3.) Occasionally within the cortex the tubular epithelium had completely disappeared, with partial or complete loss of the basement membrane. Even in this interstitial location inflammatory reaction was minimal, consisting of histiocytes, lymphocytes and rarely eosinophilic leukocytes. Foreign body giant cell reaction was not seen. Erythrocytes were rarely present in collecting tubules. Occasionally in the denser scars obliteration of glomeruli was seen as well as foci of fibroblastic proliferation. (Fig. 5.) These older appearing scars also showed relatively less crystalline material than the fresher appearing lesions. There was somewhat less fat in the proximal convoluted tubules in all animals than is usually seen in normal cats [*5,6*].

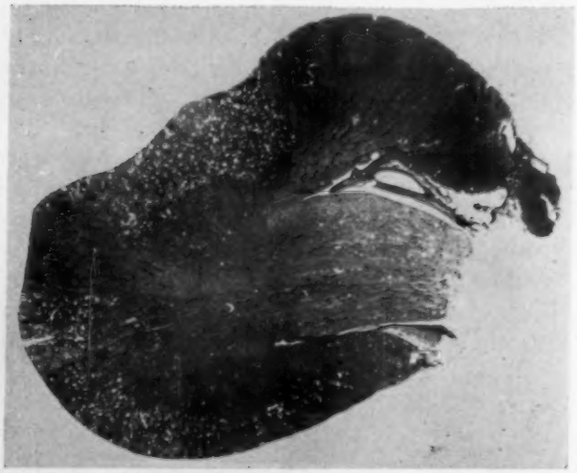

FIG. 2. Low power view of whole kidney section photographed under partially polarized light. There is focal concentration of birefringent crystals in the cortex and to a lesser extent in the papilla, with scattered crystals elsewhere in the medulla. Hematoxylin and eosin stain, original magnification × 4.

The crystals occurred as needles and rods arranged in symmetrical rosettes or sheaves or as irregular plates and smaller fragments. (Fig. 6.) Calcium could be readily demonstrated by the microscopically controlled application of 10 per cent sulfuric acid, resulting in solution of the crystals and slow precipitation of typical calcium sulfate crystals. The original crystals were soluble in 10 per cent hydrochloric or nitric acid but insoluble in 10 per cent or glacial acetic acid, various bases and a number of aromatic solvents. The crystals resisted incineration at 650°C. and following this procedure dissolved in acids with release of gas bubbles which produced an abundant precipitate in a solution of lead acetate. The crystals only rarely showed a weak surface staining with Harris' hematoxylin. Staining with cresyl violet or alcian blue was moderately intense. Von Kossa's stain and alizarin red S gave no color. Frequently, clusters of spherical crystalline bodies were seen which were well stained by hematoxylin or by the periodic acid-Schiff reaction. (Fig. 6.) These latter staining reactions were probably associated with the presence of protein which on occasion was seen to mask the birefringence of the underlying crystalline material in both stained and unstained preparations. When the aniline blue spot test for oxalate [*4*] was carried out directly on sections of kidney, 5μ. in thickness, there was gross production of blue dye. However, due to the accompanying maceration of the section and the solubility of the dye produced, histochemical localization was not possible. No crystalline material was seen other than that at the sites mentioned except for minute calcium spicules (not studied further) in the adrenal cortices of two of the cats receiving 2 mg. of vitamin B_6 per kilo of diet.

The bone marrow of the four cats receiving an unsupplemented diet had a normal degree of cellularity but showed a relative decrease in the number of normoblasts. Megakaryocytes were prominent in the spleen of three of these four cats and in one of the cats receiving 1 mg. of vitamin B_6 per kilo of diet, when compared to cats receiving 2 mg. of vitamin B_6 per kilo of diet. Such changes are probably within normal limits in the light of observations made on a

FIG. 3. Low power view of tip of renal papilla photographed under partially polarized light. There is abundant crystalline material in dilated terminal tubules and under the pelvic epithelium. Crystals may even be seen beneath the parietal epithelium, X. Those crystals lying free in the pelvis are artefactual and may represent dislocation of epithelialized intrapelvic deposits. (See Fig. 4.) Hematoxylin and eosin stain, original magnification × 30.

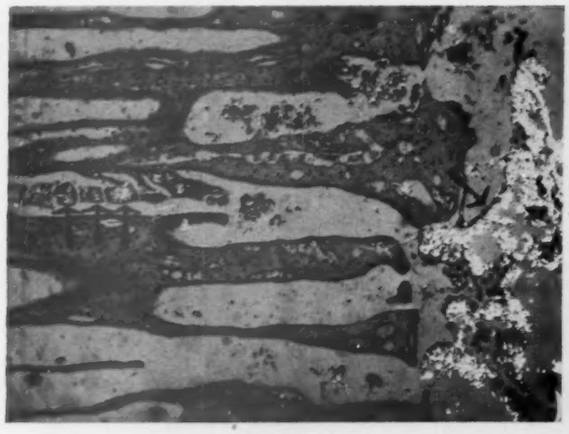

FIG. 4. Higher power view of the same kidney seen in Figure 3, photographed under partially polarized light. There is epithelialization, indicated by arrows, of some of the tubular crystals and of larger intrapelvic masses, the latter apparently attached to the tip of the papilla. Hematoxylin and eosin stain, original magnification × 60.

large number of cats fed 4 mg. of vitamin B_6 per kilo of diet. The usual degree of splenic myelopoiesis was present and there was no evidence of splenic erythropoiesis.

Hemosiderin, generally extracellular, was present in moderate amounts in the spleens of the four cats receiving an unsupplemented diet and in one of the four cats supplemented with 1 mg. of vitamin B_6 per kilo of diet. Although this pigment is inconspicuous in the reticuloendothelial system of normal cats, one of the cats given a diet supplemented with 2 mg. of vitamin B_6 per kilo showed hemosiderin deposition comparable to that described. In four animals there were large amounts of finely divided hemosiderin within the liver cells of the central areas, sufficient to give the gross liver a bright orange color.

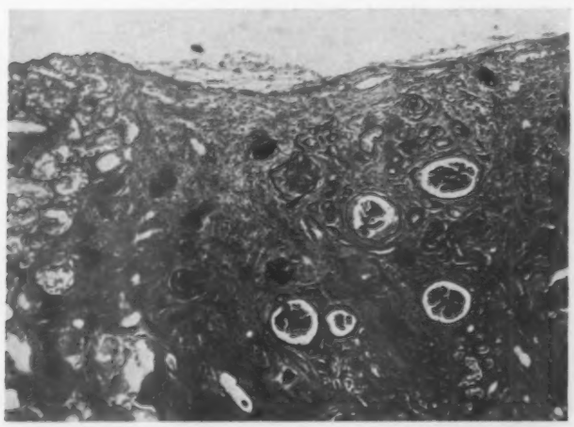

FIG. 5. High power view of edge of old cortical scar showing thickening of glomerular and tubular basement membranes, hyperplastic thickening of arterioles, obliteration of occasional glomeruli, and a definite increase in fibrous tissue. Periodic acid-Schiff stain, original magnification × 60.

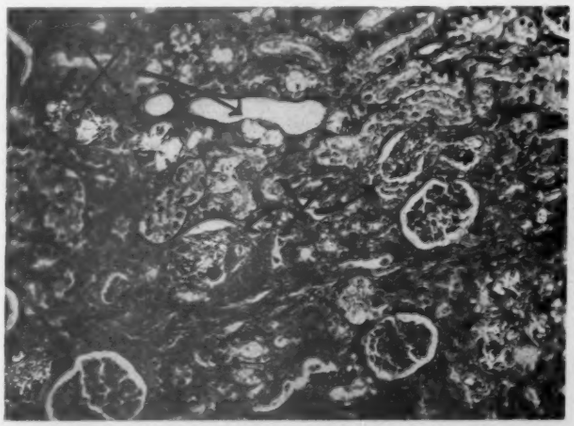

FIG. 6. High power view of crystalline deposits in cortical tubules, photographed under partially polarized light. Birefringent plates and rods are arranged in sheaves and rosettes, X, with varying degrees of atrophy of tubular epithelium and little or no epithelialization of crystals. In addition, several amorphous aggregates of crystalline material can be seen with minimal degree of birefringence here masked by hematoxylin staining, Y. Hematoxylin and eosin stain, original magnification × 120.

Two of these animals were fed an unsupplemented diet and two were receiving 1 mg. supplements of vitamin B_6 per kilo of diet. In general, the deposition of hemosiderin appeared to be associated with the presence of anemia, although two of the cats fed an unsupplemented diet failed to show hemosiderin in the liver. There was no significant increase in hemosiderin in the lymph nodes or bone marrow. One of the cats given an unsupplemented diet did, however, have extensive deposition of hemosiderin in the duodenal villi as well as in a lymph node (which in addition showed an unidentified fungus).

Stainable liver cell fat was variable, and while several of the cats receiving 1 mg. of vitamin B_6 per kilo of diet showed increased fat as compared with those receiving larger amounts of vitamin B_6, two of the animals receiving an unsupplemented diet showed no liver cell fat.

No cellular or myelin changes could be seen in the brain, spinal cord, or peripheral nerves. Gross Sudan stains of hearts and aortas were negative and no microscopic changes could be detected in the aortas or visceral arteries. Incidental pathologic changes included scattered granulomatous inflammatory foci in the myocardium, tongue and pancreas of a cat receiving 1 mg. of vitamin B_6 per kilo of diet, and organizing pulmonary arterial and venous thrombi in one of the cats given an unsupplemented diet.

COMMENTS

There was a great interest in the metabolism of oxalic acid during the last half of the nineteenth century and the first third of this one, but latterly the subject has been rather neglected by biochemists. Oxalic acid is metabolically inert in the rat, being neither oxidized nor converted to glyoxylic acid [*7,8*]. It is usually synthesized in small quantities by animals from glyoxylic acid which may be derived from glycine [*9*] or it may be found as a degradation product of ascorbic acid [*8,10,11*]. It has been demonstrated that small amounts of oxalic acid are formed in guinea pigs from glucose [*10*], and in certain microorganisms oxalic acid is produced from glucose via pentoses and by the splitting of oxalacetate [*12*]. Urinary oxalic acid in human beings (normally 10 to 40 mg. per day) is generally believed to be derived mainly from oxalates in foods and to a smaller extent from endogenous metabolism. The source of the oxalic acid measured in this study is as yet undetermined. If it is derived from glyoxylic acid, it may in large part result from decreased activity of a transaminase system such as that described by Cammarata and Cohen [*13*], i.e., glycine + α ketoglutaric acid $\leftrightarrow$ glyoxylic acid + glutamic acid.

Human diseases associated with oxalic acid are of considerable importance. Prien [*14*] in a study of 1,000 urinary calculi found that 67 per cent were either pure calcium oxalate or calcium oxalate plus apatite. Bell [*15*], in a study of approximately 25,000 autopsies, reported the incidence of renal calculi as 1.12 per cent, 0.38 per cent of the deaths being related directly to the presence of calculi.

The remarkable epidemic wave of cases of oxalate urinary stones which occurred in Central Europe after the first World War has never been adequately explained, although it was generally attributed to nutritional factors [*16*]. The report of oxaluria in British troops in India indicated that this condition may be accompanied by renal colic, hematuria or epididymitis in the absence of demonstrable stone formation [*17*].

There are, to our knowledge, twenty-two published cases of idiopathic oxalosis in man confirmed by histologic studies [*18–20*]. The majority of these patients have been adolescent males with a long-standing history of repeated episodes of calcium oxalate nephrolithiasis, in some instances complicated by pyelonephritis. The prognosis in such cases is grave, death usually occurring through renal failure and uremia. The disease is attributed to an error in metabolism leading to an accumulation of oxalates [*21*] and in some authenticated instances there is a familial distribution [*20*].

The histologic findings in these cases are very similar to the experimental findings in the animals of this study. The inflammatory response in the cat is generally of the same low grade as in man, although foreign body giant cell reaction is conspicuously absent. The relative paucity of interstitial fibrosis and the absence of gross stone formation common in man might well have been due to the relatively short duration of this experiment. The presence of crystals beneath the pelvic epithelium in several of the cats could have supplied a nucleus for future gross stone formation. The greatest difference between the experimental animals and patients with oxalosis was the absence of extrarenal oxalate deposits in the former. Such extrarenal human sites have included bone, heart, spleen, liver, thymus,

among others. Here, too, the discrepancy between the experimental and the human lesions may be related to the longer period of development of the latter.

The European literature of a generation ago contained numerous reports of a variety of conditions accompanying hyperoxalemia and/or hyperoxaluria, with symptoms referable to the gastrointestinal tract, the urinary tract, muscles, joints, tendons and the central nervous system. These reports have been summarized in the excellent review of Jeghers and Murphy [*18*].

Hyperoxaluria and hyperoxalemia associated with diabetes and hyperglycemia have been demonstrated by many workers in human subjects and experimental animals [*22,23*]. Interestingly, diabetic subjects excrete more xanthurenic acid than non-diabetic subjects [*24*]. The high xanthurenic acid excretion of diabetic subjects following a tryptophan load is markedly reduced by pyridoxine administration [*25*]. Elevation of serum and urinary oxalate has been reported in a number of other diseases, such as renal insufficiency, cirrhosis, cardiac failure and nutritional deficiencies [*18*].

The clinical picture of acute oxalate poisoning, if the patient survives the first twenty-four hours, may be that of renal insufficiency with oliguria and albuminuria [*26,27*], proceeding to anuria and death. Recovery is heralded by polyuria [*18*]. Intracellular oxalate crystals within the renal tubular epithelium have been described at autopsy in a case of oxalate poisoning [*27*].

The literature on experimental oxalate-induced urinary tract lesions has been reviewed by Dunn et al. [*28,29*] and by Jeghers and Murphy [*18*]. Such renal lesions consist primarily of necrosis of the epithelium of the proximal convoluted tubules and deposition of calcium oxalate crystals. Although early writers considered crystal deposition primary, more recent reports [*28,30*] have described tubular epithelial changes as antedating crystal deposition, the latter being relegated to a secondary passive role both as to tissue reaction and renal physiology. The long term nature of the lesions herein reported probably precludes any decision as to the primary change, although it should be added that tubular necrosis was not encountered in the absence of crystals and the latter were on occasion seen within normal-appearing tubules. Several authors have pointed out that at the stage of crystal deposition, polyuria rather than oliguria occurs [*26,28*]. In the present material, tubular dilatation was surprisingly inconspicuous as compared with that in other forms of experimental renal tubular crystal deposition (unpublished data).

Although the glomeruli are generally considered to be uninvolved, several authors have mentioned early changes therein [*28,31*]. In the latter report transitory glomerular stasis is described, followed by hemorrhage from the glomerular tufts, and the authors also stress the finding of generalized renal avascularity. Red blood cells within renal tubules were seen in only one instance in the present study. We are unaware of any oxalate-induced experimental lesions with gross scarring such as produced by vitamin B_6 deficiency in cats, although Ebstein and Nicolaier do illustrate comparable lesions produced by feeding oxalic acid and oxamide in dogs [*32*]. Renal calculi have been produced in only a few instances, in dogs and rabbits by feeding oxalate combined with oxamide or calcium chloride [*33,34*], and in dogs fed sodium oxalate [*35*].

Birch [*36*] and Cerecedo and Foy [*37*] have reported hematuria in rats deficient in vitamin B_6 and one of Birch's animals had a ureter blocked by a calcium stone. Agnew [*38,39*] has reported hematuria and renal hypertrophy in rats deficient in vitamin B_6. He also found macroscopic scarring and pitting of the kidneys in hooded rats deficient in vitamin B_6 but not in albino rats. Marked deposition of a calcium-containing compound was often seen in the kidneys of hooded rats deficient in vitamin B_6. Inasmuch as calcium deposition occurred in many of his stock and control rats, this observation was not pursued.

In work to be reported from this laboratory, endogenous oxalate production accompanied by urinary tract calculi of calcium oxalate monohydrate with obstructive sequellae similar to those seen in human beings have been consistently observed in rats deficient in vitamin B_6. The renal alterations described by Agnew may well have been related to oxalate synthesis. It should be pointed out that there was never any suggestion of hematuria in the cats deficient in vitamin B_6, and benzidine tests performed at various intervals during the experiments were negative.

As observed in other species, a marked degree of anemia occurred in the cats deficient in vitamin B_6. However, the histologic appearance of

the marrow differed from previous reports. Unlike the hyperplasia seen in the rhesus monkey [*40,41*], there was a relative deficiency of normoblasts in the cat marrow. In contrast to the distribution of the hemosiderin deposits seen in swine [*42*], the hepatic cells of the deficient cats showed preponderant involvement.

Although frequent convulsions were observed in this study, histologic changes in the nervous system were absent. Demyelinization and nerve cell changes have been reported in swine deficient in vitamin B_6 after three to four months on the diet [*43*]. The skin changes described in other species were absent [*40,44–46*]. Fatty livers comparable to those seen in rats [*47*], mice [*48*] and swine [*42*] were not encountered, nor were there the adrenal and lymphoid tissue changes reported in the rat [*38,49,50*].

Snell [*51*] has suggested that since the enzymes associated with sulfur metabolism are sensitive to vitamin B_6 deficiency, deficient animals might excrete less sulfate than well nourished ones. The data shown in Table 1 appear to support this idea. Da Silva et al. [*52*] have reported that cats deficient in vitamin B_6 do not excrete xanthurenic acid. We have confirmed this. In a private communication Da Silva has noted epileptic fits, occasional microcytic anemia, kidney lesions and iron deposition in the liver in his cats deficient in vitamin B_6.

Until recently, pathologic conditions in man due to vitamin B_6 deficiency were unknown, probably because of the widespread occurrence of this vitamin in food. There is evidence that in the processing of foods much of the vitamin B_6 may be inactivated. In the years 1951 to 1953 a commercial milk formula resulted in the production of a vitamin B_6 deficiency state in a number of infants [*53–55*]. In this laboratory renal lesions indistinguishable from those of vitamin B_6 deficiency have been observed in cats fed a commercial cat food containing more than 4 mg. of vitamin B_6 per kilo on a dry weight basis as determined microbiologically following acid hydrolysis. The crystalline material in the kidneys of these cats was identified by Drs. E. L. Prien and C. Frondel by x-ray diffraction techniques as calcium oxalate monohydrate. These results will be published elsewhere.

There is increasing evidence that the vitamin B_6 requirements of some persons may be considerably increased. Hunt et al. [*56*] and Bessey et al. [*57*] have reported cases of vitamin B_6 dependency in convulsive infants who required large doses of vitamin B_6. Schreiner et al. [*58*] indicate that some patients with seborrheic dermatitis have a metabolic defect which results in an increased local requirement for vitamin B_6 in their skin. There is also evidence that often in pregnancy the requirement for vitamin B_6 may not be met [*59–62*].

As far as we are aware, this is the first study to relate endogenous oxalate production to vitamin B_6 nutriture. The possible applications of the findings to a variety of human disease states in which oxalate plays a role, particularly those of renal stone formation and oxalosis, are obvious and warrant investigation.

SUMMARY

Vitamin B_6 deficiency, characterized by failure of growth, emaciation, convulsions, anemia and oxalate nephrocalcinosis, has been produced in cats.

The presence of large quantities of oxalate in the kidneys and urine of cats deficient in vitamin B_6, in amounts sufficient to cause marked renal damage, is attributable to excessive endogenous formation of oxalate.

The implications of these findings in relation to human disease states associated with oxalate are discussed.

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Review

Erythrocyte Cholinesterase Titers in Hematologic Disease States*

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PRIOR to 1940 very little was known about the cholinesterase of erythrocytes beyond the fact that the presence of the enzyme was readily demonstrated and that there were wide species differences in its activity. In that year Alles and Hawes [*1*] published the results of an investigation which showed that the cholinesterase of erythrocytes was qualitatively different from that of serum and plasma. This was evidenced by different behavior toward acetylcholine and several of its methyl derivatives, and by different responses to pH and salt concentration.

At about the same time the accidental discovery that large increases in erythrocyte cholinesterase activity occurred in dogs following phlebotomy led me to an investigation of anemias in man. As in the dogs, titers far above the normal range were found to occur in patients recovering from hemorrhage. In pernicious anemia a consistent pattern of events was found: In relapse, the titer was low; with the reticulocyte response to injected liver extract the values rose sharply, reaching levels far above normal; a high level was maintained during the early weeks of treatment and then the titers gradually fell to normal as remission was established [*2*]. These findings have been confirmed by a number of investigators [*3–7*]. A pattern similar to that in Addisonian pernicious anemia was reported in the macrocytic anemia of pregnancy by Cline, Johnson and Johnson [*5*] and by Pritchard and Weisman [*8*], and in non-tropical sprue by Scudamore, Vorhaus and Kark [*7*], who also found increases in the cholinesterase titers with reticulocyte responses in hypoplastic anemias. The inference from all these observations is that increased cholinesterase titers in the erythrocytes of the peripheral blood reflect accelerated erythropoiesis.

Grob and his associates [*3*] showed that administration of diisopropyl fluorophosphate in doses sufficient to suppress the high cholinesterase titer in pernicious anemia during treatment did not retard the process of remission, therefore that the high titer is a sign of recovery but not a part of the mechanism.

In 1949 Pritchard [*9*] published the first direct experimental evidence that high cholinesterase activity is characteristic of young erythrocytes. By centrifugation he separated the cells of rats subjected to blood loss into "reticulocyte-rich" and "reticulocyte-poor" fractions and demonstrated that the former had a high and the latter a low cholinesterase titer. Using Pritchard's procedure, Sabine [*10*] obtained the same results in the blood of a normal subject and in the blood of patients with reticulocyte responses. The contrast was more marked in the latter.

Recent work of Borun, Figueroa and Perry [*11*] provides confirmation that erythrocytes are well separated into younger and older fractions by centrifugation. They showed that metabolized radioactive iron appears first in the cells which accumulate in the upper layer and later in the middle and bottom layers.

In 1955 Allison and Burn [*12*] showed that young erythrocytes contain not only more cholinesterase but also more catalase and glyoxalase than do older ones. The authors used blood with a high reticulocyte count from patients with pernicious anemia of the Addisonian type, pernicious anemia of pregnancy and

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anemia from postpartum hemorrhage. They also investigated the survival of catalase and cholinesterase activity in cells transfused from O-type donors to A-type recipients and showed that the loss of activity of both enzymes with time was compatible with an exponential decay curve, assuming that in a hematologically normal individual cells of all ages from zero to 120 days are present in equal numbers. They estimated the activity of new cells as approximately three times that of the total population.

Barnard and his associates have published numerous papers on cholinesterase titers in relation to many clinical conditions, including blood dyscrasias. Their interest has centered around possible cholinergic mechanisms for clinical manifestations of disease [*13–17*]. Recently Barnard has expressed his belief in the validity of the erythrocyte cholinesterase titer as an indicator of erythropoietic activity [*18*].

In some of the more complex hematologic disorders the results are puzzling. Sawitsky and co-workers [*6*] reported sporadic observations on a large number of patients with blood dyscrasias. Their conclusions are based on the mean values for each condition regardless of the spread in the individual values, which was very wide. For example, in chronic myelocytic leukemia the mean value was approximately normal but the spread was from 56 to 150 per cent of the normal mean. The results in Hodgkin's disease and polycythemia vera were similar to those in myelocytic leukemia. In acute leukemia and "myelophthisic" anemia low values predominated and in chronic lymphocytic leukemia there was a tendency toward high values although twenty-two of twenty-four were within the normal range while only two were significantly high.

High cholinesterase titers are frequently seen in the absence of reticulocytosis. This finding is readily explained by the persistence of a high concentration of the enzyme after the stainable reticulum has disappeared.

Scudamore et al. [*7*] conclude that "it is possible that the erythrocyte cholinesterase activity may be a more accurate index of hematopoiesis than the reticulocyte count." These authors properly remark on the need for more data in hemolytic anemia and some explanation of the variable results of leukemias.

The present study deals with some of the questions raised by other investigators but chief emphasis will be placed on those areas in which interpretation of the erythrocyte cholinesterase titer would seem to be sufficiently clear for clinical application.

METHODS

Two designs of clinical investigation were followed. Sporadic observations were made in miscellaneous conditions in which the peripheral blood picture was abnormal or in which a pathologic process might involve the bone marrow. Patients were selected for close study if they had diseases in which early detection of changes in the status of the erythropoietic system would presumably be of practical value or in which there was reason to suppose that the results might contribute to the diagnosis. The rather small volume of clinical material in Los Alamos was augmented by the shipment of blood samples from Rochester, New York. Both types of investigation have been continued in San Francisco.

Blood samples for immediate use were ordinarily collected by fingerprick or heelprick with dry heparin as anticoagulant. Hematocrits were determined by an accurate semi-micro method [*19*] to permit expression of titers in terms of volume of red cells used. Oxalated venous blood from patients in Rochester was chilled and shipped by air in insulated mailing tubes, accompanied by clinical data, hematocrit values and reticulocyte counts. Many of the data on hemolytic disease were obtained by this means. Preliminary experiments showed that under ordinary conditions the enzyme activity would be preserved during the trip.

Cholinesterase activity was measured by a colorimetric method [*20*] in which the use of quinidine as a selective inhibitor of the activity of the plasma makes it possible to measure that of the erythrocytes in small samples of whole blood.

Cholinesterase titers (hereinafter abbreviated RBC ChE) are expressed per unit volume of erythrocytes unless otherwise stated. Some investigators prefer to express their values with respect to the hemoglobin content of the sample. With normochromic, normocytic blood, either manner of expression is satisfactory. In our experience moderate hypochromia without microcytosis is fairly common, and expression of RBC ChE of such subjects with respect to hemoglobin gives misleading high titers. A later section of this article is devoted to the subject of microcytic anemias, in which neither the volume of cells nor the hemoglobin is a good reference value.

The mean normal titer, 10.5 μM/minute/ml. RBC for Los Alamos [*20*], and 9.7 for San Francisco,*

* It is not certain whether the difference is a true one, attributable to geographic or ethnologic factors, or a technical one resulting from some obscure difference in experimental conditions. The values for patients in Rochester have been referred to the higher (Los Alamos) normal in order to avoid the possibility of erroneous accentuation of the high RBC ChE in hemolytic disease.

has been arbitrarily set equal to unity, and individual values expressed as decimal fractions of this unit. For practical purposes, the range for normal adults, children and older babies is from 0.80 to 1.20 units, with 90 per cent of normal values falling between the limits 0.85 to 1.15. The range for newborns is much lower. Jones and McCance [*21*] showed that the erythrocytes of cord blood contain smaller amounts of several enzymes including cholinesterase than do those of adults. The data of Pritchard and Weisman [*8*] for full term fetuses, and ours from birth to fifteen days [*20*], are in close agreement with those of Jones and McCance, with a mean value 67 per cent of the adult mean. The transition to adult values is made during the early months of life; scattered determinations in babies from two months of age upward have given values within the normal adult range.

RESULTS AND DISCUSSION

Sporadic determinations of RBC ChE in over 400 patients with hematologic abnormalities of all degrees of severity and complexity have resulted in the emergence of a group of disorders in which interpretation of the data is quite straightforward, another in which it is difficult or speculative, and a third in which the results are so consistently normal that there is little reason for making the determinations unless the differential diagnosis includes a condition in which an abnormal RBC ChE is commonly found. In our experience the second group has consisted of cases in which the disease process was complex and the diagnosis frequently obscure.

Repeated observations of individual patients have shown that the observation of trends, usually within the normal range, may provide very early evidence of remission or exacerbation in leukemias and multiple myeloma, and of recovery in erythroblastosis fetalis.

Hemolytic Disease

In Table 1 are shown the results of observations in ten cases of hereditary spherocytosis and two apparently normal relatives of patients with the disease, in one case of thalassemia major and three of thalassemia minor, and in five cases of acquired hemolytic disease not associated with known underlying disease.

Hereditary Spherocytosis. The data offer conclusive evidence that a high RBC ChE is characteristic of hereditary spherocytosis and persists to a greater or lesser degree after splenectomy even when a complete clinical cure has been effected. The finding of RBC ChE 2 to 3 standard deviations above the normal mean in persons splenectomized several years previously, with normal hematocrits and normal or minimally increased reticulocyte counts, suggests abnormal destruction of erythrocytes of a degree so slight as not to be readily demonstrated by methods in common use, with corresponding erythropoietic compensation. Of the three patients with higher titers, two (M. O'C. and E. S.) were in the third trimester of pregnancy and two

TABLE 1
HEMOLYTIC DISEASE

Diagnosis and Clinical Data	Packed Cell Volume (%)	Reticulocytes (%)	Red Blood Cell Cholinesterase (Units)*
Hereditary Spherocytosis			
Not splenectomized:			
J. O'C., 4 yr., male, child of M. O'C.	38	2.4†	1.36
J. D., 37 yr., male	37	5.5†	1.21
S. K., 33 yr., female	34	5.8†	1.28
Pre- and postoperatively:			
E. S., 29 yr., female, preoperative	37.5	1.4	1.44
1 wk. after splenectomy	37.5	.....	1.53
9 mo. later, 8 mo. pregnant	46.5	.....	1.60
Splenectomized years previously:			
M. O'C., 37 yr., female, mother of J. O'C., 7 mo. pregnant	37	2.8†	2.08
E. D., 12 yr., male	38	0.1†	1.21
J. V., 17 yr., male	37	1.2†	1.54
H. E., 16 yr., female	43	1.5†	1.21
F. C., 47 yr., male	43	1.5†	1.25
G. C., 16 yr., male	45	2.7†	1.19
Apparently Normal			
Child of E. S.	42	.....	1.24
Father of J. V.	45	1.9†	1.29
Thalassemia Major			
J. T., 35 yr., male, terminal; bone structure virtually replaced with erythroid tissue	19	9.7	2.29
Thalassemia Minor			
M. V., 35 yr., female	29	2.0†	0.93
M. S., 32 yr., female	38.5	1.8†	0.86
S. F., 33 yr., female	40	1.4†	0.90
Acquired Hemolytic Disease. Apparently Uncomplicated			
J. W., 54 yr., male, not splenectomized	23.5	12.5†	1.32
J. B., 87 yr., female, not splenectomized	23	20.0†	1.23
D. H., 10 yr., male, splenectomized 1 mo	31	31.0†	1.30
S. T., 60 yr., female, splenectomized 6 yr.	44	1.0†	0.96
R. M., 49 yr., female, remission on steroids	42	0.9†	1.10

* Mean normal 1.00; normal range 0.80 to 1.20 with normal distribution.

† Reticulocyte counts made in Rochester, where the figures for normal individuals are up to 2.0 per cent.

(M. O'C. and J. V.) had moderate residual anemia to provide a stimulus to erythropoietic response. In E. S., who was lost to follow-up after the third observation, the possibility cannot be ruled out that the high RBC ChE at eight months, when there was no anemia, may have been associated with a recurring hemolytic tendency not as yet manifested clinically. The only conclusion to be drawn at present is that patients with hereditary spherocytosis can have a very high RBC ChE without corresponding clinical manifestations.

The observation of an RBC ChE of 1.24 and 1.29 in two apparently entirely normal relatives of patients with frank disease suggests that these individuals may carry the trait in latent form. Both parents of J. V. were free of any evidence of the trait demonstrable by usual methods. As a result of several such observations of his own and others in the literature, Young [*22*] suggested that "one of the parents carries the gene for HS but the gene penetrance or expressivity is such as to be undetectable by available methods (? 'carrier' state)." It may be that further investigation will provide a means of accounting for apparent departures from the pattern of inheritance as a mendelian dominant in some family trees.

Thalassemia. The exceedingly high RBC ChE observed in J. T. is in keeping with the appearance of the peripheral blood smear and with a markedly shortened red blood cell life span as estimated by labeling with Cr^{51} (half life of labeled erythrocytes three to four days as compared with twenty-six to thirty days in the normal) [*23*]. The contrast between the values of RBC ChE for J. T. and for the three patients with thalassemia minor is consistent with other evidence that the difference between the two conditions is not entirely one of degree.

Acquired Hemolytic Disease. When a hemolytic tendency appeared to be the primary disorder, the RBC ChE was high when the disease was active, and normal when it was in remission. The titer, however, was not high enough to be in keeping with the very high reticulocyte counts frequently seen.

When hemolytic anemia appeared during the course of underlying disease, the RBC ChE varied from far below normal (0.57) in patient with advanced Hodgkin's disease to exceedingly high (2.2) in a patient with probable acute monocytic leukemia to be mentioned subsequently. Discussion of this problem will be more profitable when the results of simultaneous observations on cholinesterase, iron turnover and isotope labeling now in progress are available.

One case, however, illustrates a situation in which the determination of RBC ChE influenced the decision to undertake splenectomy as a last resort.

CASE 1. (*Myelosclerosis, extramedullary erythropoiesis, pancytopenia, excessive splenic sequestration and phagocytosis.*) A fifty-one year old woman was admitted because of severe anemia. A bleeding tendency had been present for eleven years and a diagnosis of myelosclerosis had been made four years prior to this admission. The hemoglobin was 2.9 gm. per cent, the hematocrit 10 per cent, the white blood cell count 1,000 per cu. mm. and the platelet count 30,000 per cu. mm. Hepatomegaly and marked splenomegaly were present. Prednisone therapy was started (10 mg., later increased to 20 mg. daily). After a period of observation and preparation with many transfusions, splenectomy was performed. The immediate postoperative course was satisfactory. The hematocrit was maintained at 36 to 38 per cent without further transfusions. The patient was discharged on the eighth day. Two weeks later she was readmitted with thrombosis of several large veins, massive infarcts and a ruptured intestine; she died of generalized peritonitis and septicemia.

The spleen weighed 1,280 gm. and histologic studies confirmed the presence of erythropoiesis and excessive sequestration and phagocytosis of erythrocytes. The liver and lymph nodes also showed many foci of erythropoietic activity.

During a nine-day interval between transfusions in the preoperative period the RBC ChE had fallen from 0.88 to 0.63 and the hematocrit from 16.5 to 10.0 per cent. The decline in RBC ChE was consistent with aging of donors' cells and no appreciable contribution of new erythrocytes from the patient's erythropoietic tissues. The rapid decrease in hematocrit could not be accounted for by blood loss.

Iron turnover studies carried out by Dr. Myron Pollycove of the Donner Laboratory indicated that although the uptake of Fe^{59} from the plasma was greatly accelerated, most of it going to spleen and liver, only 10 per cent of the injected dose appeared in the peripheral erythrocytes as compared with 85 to 100 per cent in the normal subject [*24*]. All these observations contributed to the clinical decision that splenectomy was indicated as a last resort.

Of four other patients with extramedullary erythropoiesis two, with agnogenic myeloid metaplasia, were doing well clinically with hematocrits of 34 and 38 per cent and RBC ChE of 1.40 and 1.86; the third patient, with cirrhosis of the liver, bleeding esophageal varices and myeloid metaplasia of the spleen, had a hemato-

TABLE II
MICROCYTIC ANEMIAS

Age and Sex of Patient, with Diagnosis	Hemoglobin (gm. %)	Packed Cell Volume (%)	MCHC (%)	MCV (cu. μ)	Red Blood Cell Cholinesterase (Units)	
					ChE/Vol.	ChE/RBC
11 mo., male, severe dietary deficiency (Case VI) untreated	3.8	20	19	42	1.18	0.56
3 wk. improved diet plus oral iron supplement	10.2	35	30	69	1.41	1.12
15 mo., female, severe dietary deficiency	4.9	21	24	45	1.37	0.67
1 yr., male, severe dietary deficiency	7.0	33	22	56	1.27	0.79
45 yr., female, iron deficiency, metrorrhagia	3.6	17	22	59	1.54	1.00
40 yr., female, iron deficiency	7.8	31	25	61	1.51	1.01
6 mo., male, congenital heart disease	12.6	41	30	62	1.42	1.01
37 yr., female, iron deficiency	8.5	33	26	66	1.39	1.00
45 yr., female, idiopathic microcytic hypochromic anemia with Plummer-Vinson syndrome*	7.2	35	21	71	1.27	0.98
44 yr., female, chronic rheumatic disease*	7.6	32	24	71	1.55	1.27
1 yr., male, iron deficiency	8.9	34	27	75	1.20	1.03
11 mo., male, etiology uncertain, not attributed to dietary deficiency	5.4	26	21	..	1.47	....
14 yr., male, non-thrombocytopenic purpura, hematuria, no response to blood loss (D. G., cf. also Table III)	13.1	36	37	72	0.82	0.66

NOTE: MCHC = mean corpuscular hemoglobin concentration. MCV = mean corpuscular volume. ChE/Vol. = cholinesterase titer per unit volume of erythrocytes. ChE/RBC = cholinesterase titer per cell. The normal range for each is 0.80 to 1.20 units.

* Case from Sabine [2].

crit of 16 per cent and RBC ChE of 0.90. The fourth case was one of probable acute monocytic leukemia with extramedullary erythropoiesis demonstrated at autopsy in the liver, lymph nodes and dura, but not in the spleen. Iron turnover studies indicated a six- to sevenfold increase in the production of erythrocytes, 63 per cent of which found their way into the peripheral blood where their life span was approximately thirteen days [24]. The RBC ChE remained constant at 2.38 ± 0.1 while the hematocrit was maintained at 35 to 38 per cent during the last four months of the patient's life.

Reticulocytosis without High RBC ChE. Occasionally, thus far only when the disease process was severe or complex, or both, and a proved or suspected hemolytic tendency was present, normal or even low RBC ChE has been observed when the reticulocyte count was increased. Pritchard and Weisman [8] reported this in two cases of hemolytic crisis in eclampsia and three of sickle cell anemia and sickle cell disease-thalassemia complicating pregnancy. We have observed it in a total of eleven patients, all with malignant hematologic disorders. In one, a patient with chronic myelocytic leukemia, a persistent reticulocyte count of 5 to 8 per cent and RBC ChE 0.67, observations were made on the top layer of the centrifuged cell pack. The reticulocyte count was 18.6 per cent, the RBC

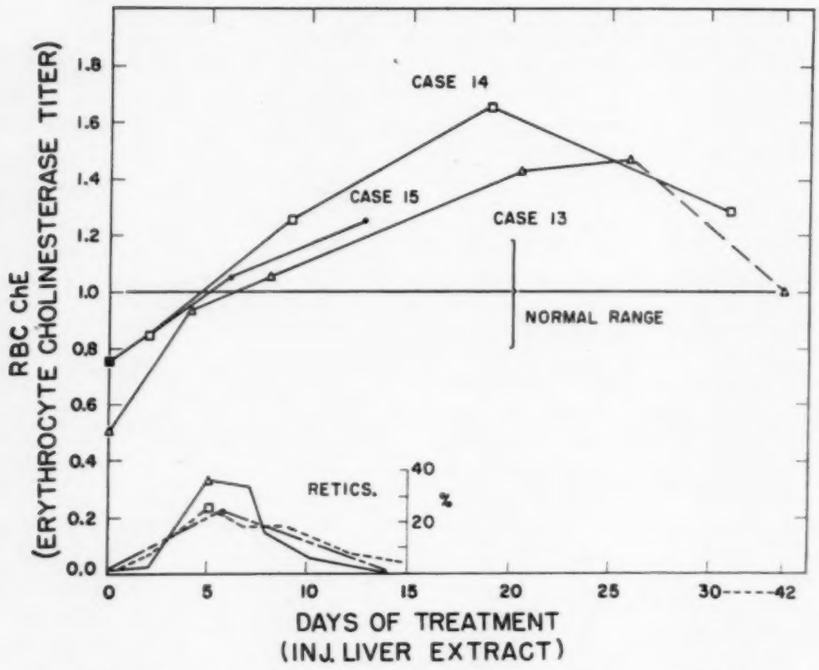

FIG. 1. Pernicious anemia. Erythrocyte cholinesterase titers (RBC ChE) and reticulocyte count (RETICS.) in relapse (zero day) and during the first month of treatment. (Adapted, with the permission of the *Journal of Clinical Investigation*, form Sabine [2], Table II.) RBC ChE, originally in terms of NaOH required to neutralize acetic acid formed, has been re-expressed as fractions of the mean titer of the control group to conform with the usage which has since been adopted.

ChE 0.83. If the reticulocytes had the normal activity of approximately 3.0,* the activity of all of the remaining cells would be 0.33.

Because of the generally poor correlation between RBC ChE and reticulocyte count, Pritchard and Weisman [8] concluded that this parameter was not a satisfactory index of erythropoietic activity. Alternatively, it is suggested that the demonstration of an abnormality in the reticulocytes may well be a means of identifying disorderly or pathologic reticulocytosis mentioned by Wintrobe [25] (p. 106).

Non-hemolytic Icterus. In obstructive jaundice and in hepatitis normal RBC ChE has been found consistently. In one case, however, a high titer was found and was accounted for by intermittent gastrointestinal bleeding. (Table IV.)

A normal RBC ChE in proved hemolytic anemia suggests underlying disease, and an elevated titer in proved non-hemolytic icterus suggests blood loss.

* Results obtained since this article went to press have confirmed by direct observation the value 3.0 suggested by Allison and Burn [12] for the activity of new red cells, and are incorporated into a forthcoming article.

Clinically Significant Trends in RBC ChE in Patients with Hematologic Disease

In order to illustrate the magnitude and time relations of trends in RBC ChE occurring with large bursts of erythropoietic activity, the data of Table II from Sabine [2] have been re-expressed as fractions of the mean normal value to conform with the usage in this paper, and plotted in Figure 1 to show the course of events in the treatment of pernicious anemia. In this disease the response to therapy can be evaluated by observations of RBC ChE at weekly intervals, if desired, without risk of missing the peak.

Similar upward trends of smaller magnitude, and generally within the normal range, have preceded or accompanied the earliest evidence of remission in leukemia and multiple myeloma, and of recovery in erythroblastosis. Rapid downward trends have preceded or accompanied exacerbations of acute leukemia and have been a frequent although not invariable feature of the terminal period.

Four representative cases have been selected to illustrate the correlations which have been

observed between the trends of RBC ChE and the short term prognosis. Others will be mentioned briefly

CASE II. (*Multiple myeloma.*) A forty-nine year old man, with a diagnosis of multiple myeloma documented in January 1952 by bone marrow aspiration, has had a number of exacerbations followed by remission, one of which, occurring in 1954–1955, is illustrated in Figure 2. Therapy had included the administration of urethane (which was discontinued shortly after admission), testosterone and multiple vitamins. Various palliative medications and supportive measures were employed during December and January. The remission which appeared in February became clinically complete and for over a year the only positive laboratory finding was an abnormal serum globulin fraction. At present, six years after the diagnosis was made, the patient has recovered from another exacerbation of the disease and is in remission, able to work full time in a responsible administrative position.

The point of greatest interest in connection with the investigation of this case was obtained in late January. The patient was febrile, icteric and critically ill with a marked hemolytic tendency. The RBC ChE, however, had risen from low normal values to the upper limit of the normal range. This was the earliest sign of remission. The maximum reticulocyte count was 2.8 per cent early in February.

CASE III. (*Acute lymphocytic leukemia.*) A four year old boy was apparently in good health until the date of admission, when he was hospitalized because of severe abdominal pain. A provisional diagnosis of acute lymphocytic leukemia was confirmed by examination of the bone marrow, in which microblastic forms predominated. The salient features of the clinical course, the therapeutic measures and the hematologic data are shown in Figure 3. It will be noted that clinical remission lasted for thirteen consecutive months.

As in Case II, a marked increase in RBC ChE was observed as one of the earliest signs of remission. When in January a hematologic exacerbation occurred, the RBC ChE remained at a high normal level, the patient was asymptomatic, chemotherapy was well tolerated and remission became apparently complete. (The bone marrow was not examined during remission.) In April a sharp decline in RBC ChE was the first sign of relapse. Methotrexate was administered when at the end of April abnormal cells reappeared in the smear and lymphadenopathy recurred. A new level of RBC ChE around

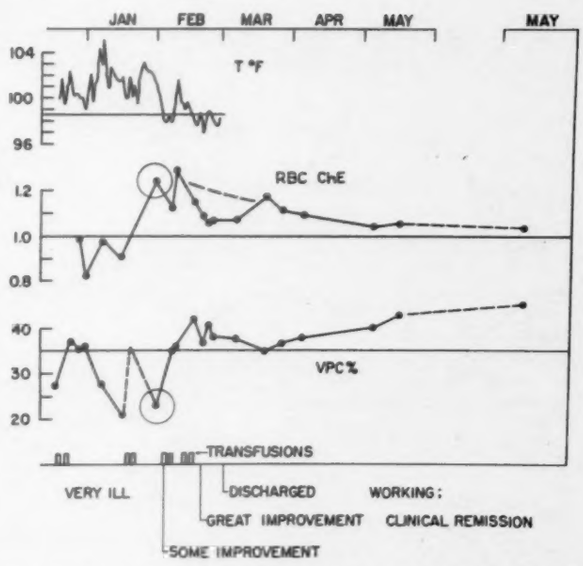

FIG. 2. Case II. Multiple myeloma. The salient features of the clinical course, the daily maximum rectal temperature (T°F), the erythrocyte cholinesterase titer (RBC ChE) and the hematocrit (VPC %) are shown from the beginning of a severe exacerbation of the disease to complete remission.

1.0 was established. In August signs of lassitude and a falling hematocrit and RBC ChE were considered indications for transfusions, and in September a sharp decrease in RBC ChE preceded a hematologic flare-up. During the last four months of life there was a steady downward trend in RBC ChE, which was masked by the addition of donors' cells with normal titers. Estimates based on the titers of the various donors indicated that the patient's own cells must have had a very low titer while the donor cells decreased in activity in a manner similar to that observed in transfused cells by Allison and Burn [*12*]. During this time, although the good response to chemotherapy continued and clinical remission was maintained, the steady downward trend in RBC ChE was regarded as an unfavorable sign and the patient was watched more closely than his clinical and hematologic status appeared to demand.

Throughout the course of the disease, after the initial stormy period until the brief terminal episode, the patient appeared to be so well that there was nothing but recurrence of lymphadenopathy and the blood picture to guide the management of the case. In this instance the determination of RBC ChE was an important and useful part of the blood picture, giving reassurance or warning which on all occasions was borne out by subsequent events.

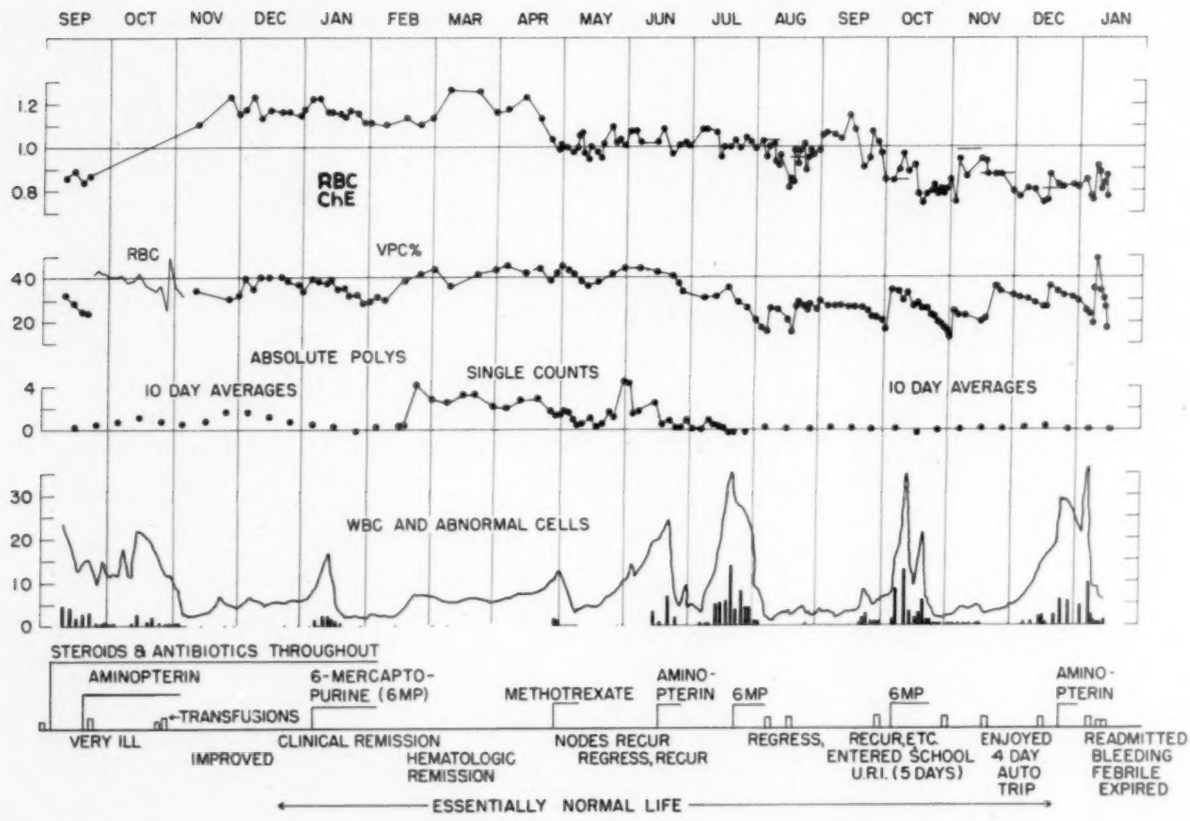

FIG. 3. Case III. Acute lymphocytic leukemia in a child. The salient features of the clinical course, the erythrocyte cholinesterase titers (RBC ChE) and the hematologic data are shown for the duration of the disease after the diagnosis was made. Total white cell counts (WBC) in thousands per cu. mm. and abnormal cell count are represented by a continuous curve and by vertical bars, respectively. When the baseline is omitted, no abnormal cells were seen. The polymorphonuclear cell counts (ABSOLUTE POLYS) in thousands per cu. mm., are plotted by ten-day averages when the points are not connected; points joined by a line are individual counts. When the point is plotted below the baseline, no polymorphonuclear cells were seen. The duration of each course of chemotherapy is indicated by a horizontal line.

CASE IV. (*Acute monocytic leukemia and infection.*) A forty-five year old woman was followed through nine months, from the time a diagnosis of monocytic leukemia was made until her death. As shown in Figure 4, clinical and hematologic remission appeared suddenly during recovery from staphylococcus septicemia. For over six months no abnormal cells were seen in the peripheral blood and the differential count was that of the leukocytosis of infection. Examination of the bone marrow in January showed minimal pathologic changes instead of sheets of blast forms present throughout the earlier specimen. The hemoglobin and platelets were maintained at normal levels. A recurrence of pancytopenia preceded the final exacerbation of the disease.

In this case levels of three blood enzymes (RBC ChE, serum lactic dehydrogenase and serum glutamic-oxaloacetic transaminase) were determined, with the results shown in Figure 4. At the beginning of clinical remission there was a rise in RBC ChE from below normal to the mid-normal range, accompanied by an increase in reticulocyte count from 2 to 4 per cent. In mid-July there was a small rise in reticulocyte count and the RBC ChE was 1.17. Late in July, when severe exacerbation of the disease appeared, there was a precipitous downward trend, interrupted by transfusion and immediately resumed, such as occurred in the later months of the disease in Case III. During this time the reticulocytes were frequently reported 0.0 per cent.

Both of the serum enzyme levels rose during the severe illness in November to figures well above normal. Normal values were found during remission, and terminally the titers of both enzymes swung upward, forming mirror images of the RBC ChE. A discussion of these findings would be premature, except to remark that rising or high levels of the two serum enzymes are

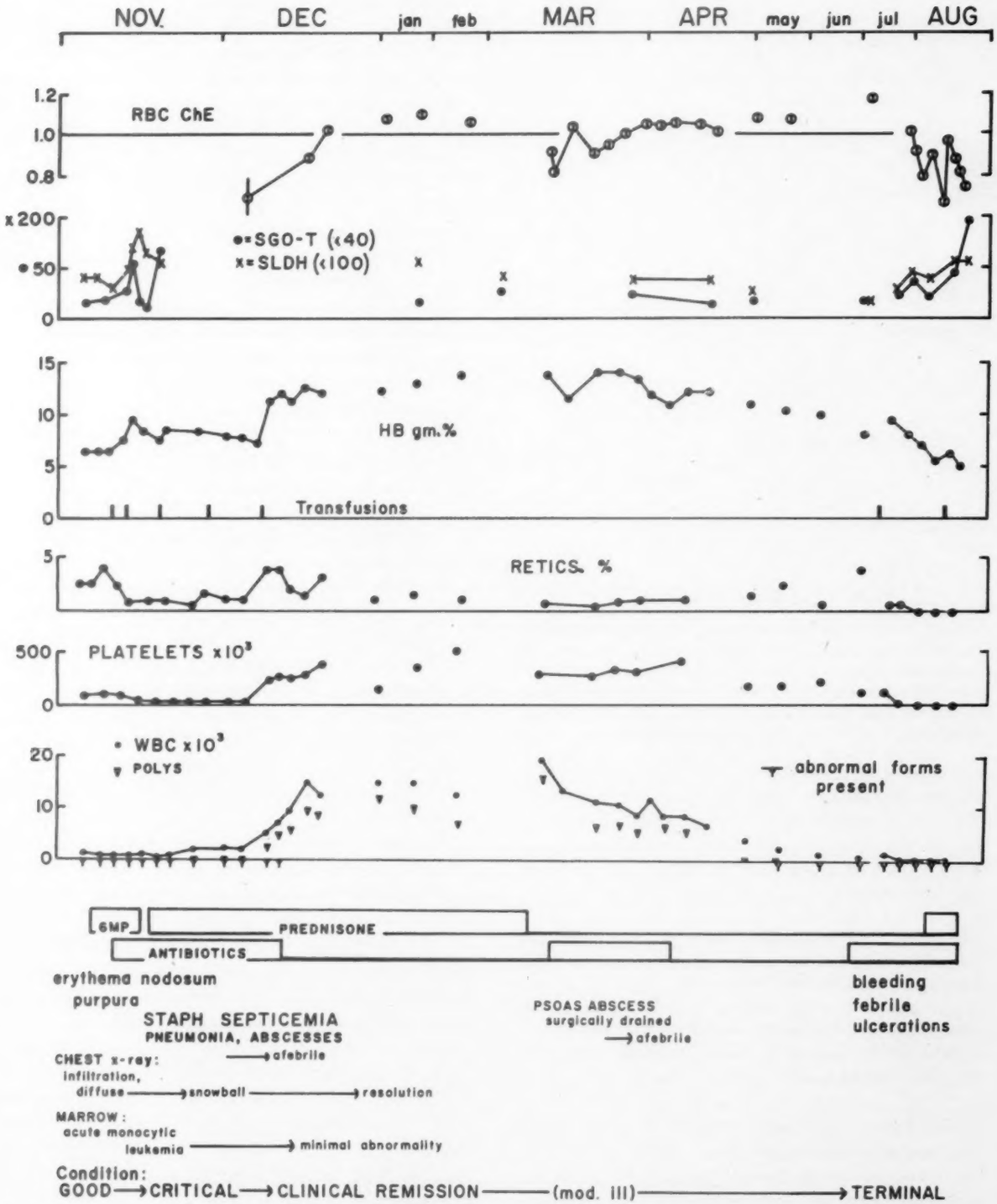

FIG. 4. Case IV. Acute monocytic leukemia with staphylococcus hemolyticus infection in an adult. The salient features of the clinical course, the hematologic data, the erythrocyte cholinesterase titers (RBC ChE) and serum transaminase-(SGO-T) and serum lactic dehydrogenase (SLDH) are shown for the duration of the disease after the diagnosis was made. Figures in parentheses indicate the maximum normal values for the two serum enzymes. On the time scale, the months during which the patient was seen in the clinic are foreshortened. Points joined by lines were obtained during hospitalizations. Because there is only one low RBC ChE at the beginning of the observations, a vertical bar has been drawn to show the range of previous determinations in acute monocytic leukemia during exacerbations.

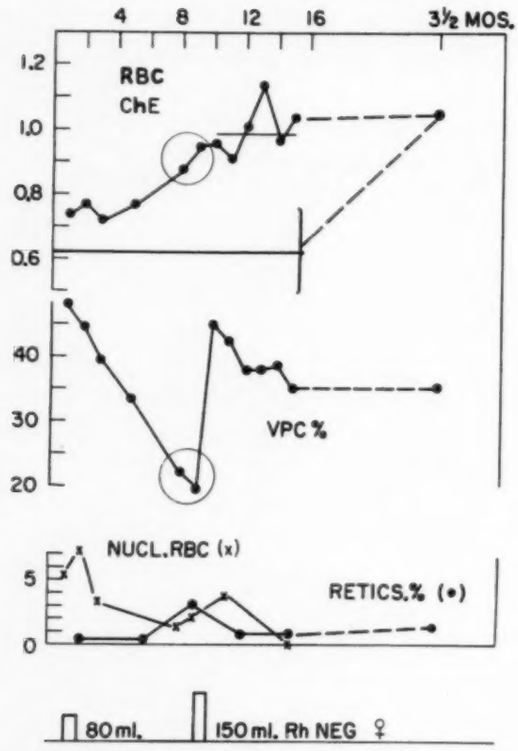

FIG. 5. Case V. Erythroblastosis fetalis. Erythrocyte cholinesterase titers (RBC ChE), hematocrit (VPC %), reticulocyte count (RETICS. %), nucleated erythrocyte count (NUCL. REC) per 100 white blood cells, and transfusions are shown for the first sixteen days of life and at three and a half months. Circles have been drawn around two pairs of values for RBC ChE and VPC %. A short horizontal line represents the calculated value for RBC ChE after the second transfusion. The range of values for normal newborn infants is indicated by a bracket.

known to be associated with unfavorable developments.

CASE V. (*Erythroblastosis fetalis.*) In this female infant, appearing normal at birth, classic signs of erythroblastosis developed within two days. She was treated conservatively, as indicated in Figure 5, and responded well clinically. When she was several months of age, moderate residual anemia and pallor were the only abnormal signs.

The points of greatest interest in this case are on the eighth and ninth days of life. The hemolytic process was unabated but the RBC ChE was well above the newborn range and the reticulocytic count was 3 per cent. After the second supportive transfusion the RBC ChE assumed an adult level approximately equal to the calculated post-transfusion value. Whether there is any connection between the rise in RBC ChE and the subsequent abatement of the hemolytic process is uncertain, although there is a similarity in this respect with the course of events in Case II. The evidence of increased erythropoietic activity was regarded as a favorable sign.

Additional Observations. A patient with multiple myeloma was observed during a brief hospitalization, and subsequently as an outpatient for four months, when the clinical remission became complete. When the patient was first seen, the hematocrit was 35 per cent and the RBC ChE 0.70. The values gradually rose to 45 per cent and 0.92, respectively.

Three patients with chronic lymphocytic leukemia were observed through a period of improvement. Two patients were asymptomatic and no therapy was given. In each the hematocrit rose from a moderately anemic level to a normal level. The RBC ChE rose from low normal to mid-normal values. The third patient was ill at the time the diagnosis was made but the situation was not critical. The hematocrit was 30 per cent and the peripheral blood was leukopenic with relative lymphocytosis. The RBC ChE was 0.80. It was decided to attempt to induce at least partial remission with steroids, and transfusions if necessary, and to delay chemotherapy until the level of RBC ChE was more satisfactory. Clinical remission occurred under this regimen without transfusion. The hematocrit gradually rose to 45 per cent and the RBC ChE to 1.0. This status has been maintained for nine months with chemotherapy still in reserve.

Probably only rarely, and never in acute leukemia, should a low or falling RBC ChE be regarded as a contraindication to chemotherapy. In chronic lymphocytic leukemia, in which there is disagreement as to the management of choice in "fresh" cases [*26*], the determination of RBC ChE deserves consideration along with all other available criteria.

Microcytic Anemia

In microcytic anemias the interpretation of RBC ChE may be misleading if the red cell size is not taken into consideration. The relevant clinical and hematologic data for twelve cases are given in Table II. The common finding in iron deficiency is a normal enzyme content of the small cells, so that the titer per unit volume of cells is high. In anemia due to marked dietary deficiency, illustrated by Case VI, the enzyme activity per cell may be reduced as it is in untreated pernicious anemia. The last case

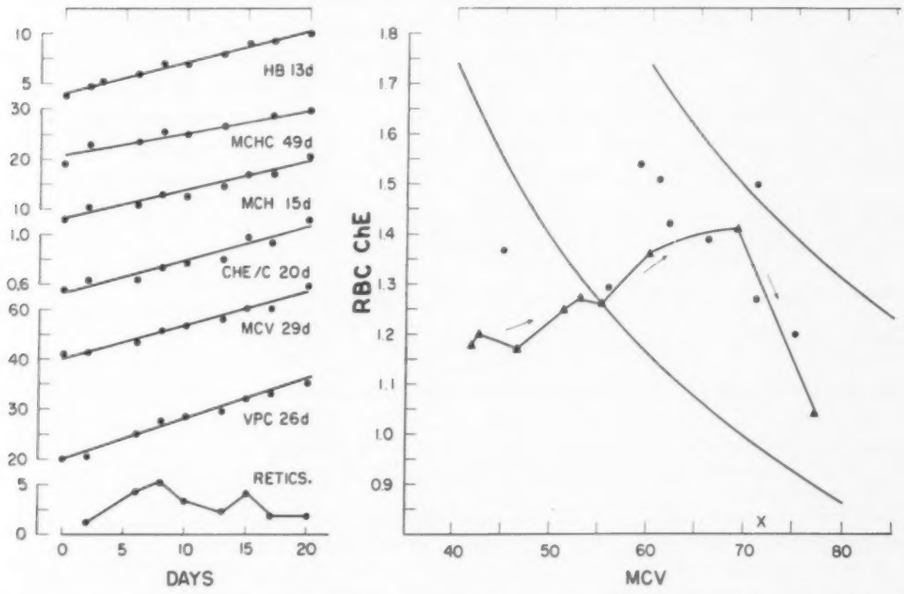

FIG. 6. Microcytic anemia. On the left is shown the hematologic progress of the patient (Case VI) during twenty days of dietary therapy. The number of days required to double each initial value is indicated. (Abbreviations: HB = hemoglobin in gm. per cent. MCHC = mean corpuscular hemoglobin in per cent. MCH = mean corpuscular hemoglobin in $\mu\mu$g. CHE/C = erythrocyte cholinesterase titer expressed per cell. MCV = mean corpuscular volume in cu. μ VPC = hematocrit. RETICS = reticulocyte count in per cent.) On the right, the cholinesterase titer expressed per unit volume of cells (RBC ChE) is plotted against MCV. Isolated symbols (•) represent nine cases of iron deficiency anemia from Table II; —▲— represents Case VI during twenty days of hospitalization and again after six weeks; arrows indicate the passage of time. The symbol (X) represents a case of purpura (D. G., cf. Tables II and III) with microcytic, normochromic anemia, discussed in the section on thrombocytopenia and purpura. The two smooth curves represent the calculated limits of RBC ChE (expressed in the usual way *per unit volume of cells*) if the activity *per cell* is within normal limits.

(D. G.), also with subnormal enzyme activity per cell, is mentioned in the section on thrombocytopenia and purpura.

CASE VI. (*Extremely severe microcytic, hypochromic anemia.*) An eleven month old boy, admitted with a complaint of persistent vomiting, weighed 12 pounds and exhibited extreme weakness and waxy pallor. He had been fed from birth on a milk formula with no solid food and no supplements. The hematologic data on admission are given in Table II. Since he was alert and not in any apparent distress unless held upright, it was decided to give him a trial on dietary therapy. Solid foods high in protein, fruit juice and an iron supplement were given. All components of the erythroid hematologic picture improved rapidly. Clinical improvement was marked. The patient was discharged. Upon his return in six weeks the hemoglobin was 13.5 gm. per cent, the hematocrit 42 per cent and the mean corpuscular volume 77 cu. μ.

In Figure 6 the left-hand portion shows the hematologic data in Case VI during twenty days of dietary therapy. The hemoglobin per unit volume of blood and the mean corpuscular hemoglobin improved most rapidly, doubling in thirteen and fifteen days, respectively. The cholinesterase activity, expressed per cell (CHE/C), doubled its initial value in twenty days. The other components improved somewhat more slowly. There was a reticulocyte response with a maximum of 5 per cent on the eighth day.

The right-hand portion of Figure 6 provides a simple nomogram for comparison of any case of microcytic anemia with the usual picture. Cholinesterase activity expressed in the usual way per unit volume of cells (RBC ChE) is plotted against mean corpuscular volume (MCV). The curves represent the limits of RBC ChE if the amount of enzyme per cell is within the normal range. Single values for ten cases of microcytic hypochromic anemia have been plotted. The progress in Case VI is shown with arrows to indicate the passage of time from the patient's admission to six weeks after discharge. The enzyme activity per cell was promptly restored to normal, and remained so during the slower progress from the microcytic toward the normocytic region.

TABLE III

IDIOPATHIC THROMBOCYTOPENIA AND PURPURA

Clinical Data and Diagnosis	Packed Cell Volume (%)	Reticulocytes (%)	Red Blood Cell Cholinesterase (Units)
J. T., 16 yr., female, idiopathic thrombocytopenia, not splenectomized	40.5	1.3	0.99
K. R., 44 yr., female, idiopathic thrombocytopenia, splenectomized years previously	41.5	2.1	1.08
K. B., 55 yr., male, idiopathic thrombocytopenia, splenectomized years previously	42.0	3.2	1.16
M. P., 28 yr., female, idiopathic thrombocytopenia, splenectomized years previously	43.5	1.5	1.03
M. T., 4 yr., female, idiopathic thrombocytopenia, not splenectomized; frequent epistaxis 3 wk.	23.0	4.4	1.50
G. T., 19 yr., male, idiopathic thrombocytopenia, recent onset; epistaxis, petechiae (transfusion (2 units) given in preparation for splenectomy before these determinations were made)	30.2	3.6	1.23
D. G., 14 yr., male, non-thrombocytopenic purpura of undetermined etiology; red blood cells normochromic, microcytic; no response to blood loss during 3 wk. following massive hematuria	44.5 36.0	... 0.5	0.79 (0.64) * 0.82 (0.66)

* Figures in parentheses = RBC ChE expressed per red blood cell.

Because it is frequently impracticable to treat patients such as the one in Case VI with diet alone and to study the hematologic changes closely, a digression from the main theme of this article seems justified. During the first twenty days of treatment there must have been rapid destruction of hypochromic microcytes, for while the hemoglobin and hematocrit approximately doubled, the red cell count rose only from 4.80 to 5.03 million per cu. mm. From cell diameter counts and the hematologic data a set of calculations was made from which it was possible to estimate the fraction of the original cell population surviving after twenty days. Cell diameter counts for the patient in Case VI and two normal subjects were as follows:

	Red Blood Cell Diameter (μ)								
	2	3	4	5	6	7	8	9	10
Case VI, zero day	2*	2½	10	18	42	19	6	½	0
20th day	0	0	1½	13	22	36	21½	5½	½
8th wk.	0	0	0	4	19	44	26	4	3
Normal subject (MCV 88)	0	0	0	½	12	53½	30	4	0
Normal subject (MCV 83)	0	0	0	2½	17½	59½	14	6½	0

* Unit of measure throughout, per cent.

The per cent counts for zero day and the twentieth day were converted into millions per cu. mm. and the former subtracted from the latter. There had been a loss of 1.7 million small red blood cells per cu. mm. (2 to 6 μ) and a gain of 2.0 million larger ones (7 to 10 μ). Since the gain in larger cells must all be attributed to the new population, the maximum survival of old cells was 3.1 million per cu. mm. or 65 per cent. The solution of simple equations gave values for the mean corpuscular hemoglobin concentration (MCHC), mean corpuscular hemoglobin (MCH) and mean corpuscular volume (MCV) for the new red cell population if these comprised 35 per cent of the total population. The results of the calculation are shown in the following table, as are the results when other likely values for MCHC were assigned to the new population:

MCHC (%)	New Red Blood Cells		Old Red Blood Cells
	MCH ($\mu\mu$g.)	MCH (cu. μ)	Survival (%)
36	40	112	65
35	36	102	59
32	32	94	51

The calculated values for MCV are probably all somewhat high, for poikilocytosis and extreme microcytosis had disappeared almost completely by the twentieth day, and the MCV of the old population therefore must have been increased. Characterization of the new population as normocytic and normochromic is compatible with destruction of about 40 to 45 per cent of the old cells in twenty days.

Observation in Miscellaneous Conditions

Thrombocytopenia and Purpura. In Table III are shown the data for seven cases. The first four patients, all with classic idiopathic thrombocytopenia, were not anemic and had no history of recent blood loss. The RBC ChE was normal. The fifth patient (M. T.) was admitted with a chief complaint of blood loss. There was severe anemia, a reticulocyte count of 4.4 per cent and RBC ChE 1.50. The sixth patient (G. T.) had received 2 units of blood just before the determination of RBC ChE, so that the observed value of RBC ChE was the net resultant of the patient's titer and the presumably normal titers of the donors. Undoubtedly the patient's titer was much higher than the moderately elevated observed value of 1.23. In the seventh case (D. G.) the etiology of the purpura was not known, nor was the microcytosis explained. During three weeks following massive hematuria, which reduced the hematocrit from 45 to 35 per cent, there was no increase in hematocrit, no change in mean corpuscular volume, no reticulocyte response and no change in RBC ChE. Values given in the table are the means of determinations made thrice weekly over the three-week period. The RBC ChE, lower than normal before blood loss occurred and undergoing no change, added to the impression that a complex metabolic disorder was present.

Anemia Associated with Chronic Renal Insufficiency. Observations have been made in fourteen cases of chronic renal disease with renal insufficiency. The hematocrits ranged from 21 to 33 per cent, and the RBC ChE from 0.69 to 1.12 with a median value of 0.945. Reticulocyte counts were less than 1 per cent in all cases. This series included two patients whose bone marrows were examined and observed to be hyperplastic. All the available evidence pointed to interpretation of the hyperplasia as representing arrest of erythroid maturation rather than erythropoietic hyperactivity.

Anemia Associated with Metastatic Carcinoma. The great majority of patients with metastatic carcinoma have had RBC ChE within the normal range, whether or not they were anemic. Subnormal values have been observed occasionally and appeared to be associated with cachexia rather than with extensive bony metastases. Patients with carcinomatosis may respond to blood loss with a rise in RBC ChE to high levels, as do normal subjects; our series of patients making this response is not large enough to permit any correlations with specific clinical or pathologic entities. In patients with carcinoma there is little to be gained by determinations of RBC ChE except when there is specific need for evaluation of erythropoietic response.

Hypoplastic Anemia; Unexplained Normocytic, Normochromic Anemia. In seven patients with normocytic, normochromic anemia with hypoplastic marrow and no evidence of other disease or of extramedullary erythropoiesis, the RBC ChE was low to mid-normal (0.46 to 1.06). In two cases clinically similar to these seven the bone marrow appeared to be hyperplastic. The RBC ChE was 0.96 and 1.03. The same results have been obtained in several clinically similar cases without sufficient documentation to permit a definite diagnosis. Scudamore et al. [7] reported on one patient with hypoplastic anemia who had a low RBC ChE when first observed which rose rapidly with a reticulocyte response. Low to normal titers appear to be characteristic of this group of anemias, and the finding of a high titer would suggest an alternative diagnosis.

Evaluation of Blood Loss

It is frequently difficult to estimate from the history whether or not a significant amount of blood loss has occurred. Data for three cases are given in Table IV. The first illustrates the finding of a moderately elevated RBC ChE in the presence of chronic blood loss which the patient did not regard as significant. The return to normal values after the menopause was established suggests that the diagnosis was correct. In the second case a normal hematocrit and normal RBC ChE indicate that the amount of blood actually lost was too small to elicit a compensatory erythropoietic response. This has, of course, no bearing on the clinical significance of hematuria. In the third case a high RBC ChE in a condition in which anemia is common but in

TABLE IV
BLOOD LOSS

Clinical Data and Diagnosis	Packed Cell Volume (%)	Reticulocytes (%)	Red Blood Cell Cholinesterase (Units)
J. S., 42 yr., female, apparently normal member of control series	45.0	.....	1.15
4 yr. later, apparently normal; Coombs' test negative, serum bilirubin normal, incubation hemolysis negative, family history non-contributory	44–46	0.5–1.0	1.33–1.36
2 yr. later, menopause established	44.0	.....	1.16
(Diagnosis of minimal menorrhagia made in retrospect.)			
R. M., 24 yr., male, unexplained recurrent gross hematuria for 2 yr.; normal renal function; "3-glass test" indicated the lower urinary tract as the source of bleeding	46.0	1.0	0.92
J. H., 54 yr., male, carcinoma of the stomach with liver metastases, obstructive jaundice, iliac and vena caval thromboses; terminal; bleeding duodenal ulcer discovered at autopsy	31.5	.....	1.32

which high RBC ChE is not expected was a clue to undetected blood loss.

Practical Considerations

Technically, determinations of RBC ChE are not difficult, nor are they unduly time consuming if several are performed at the same time. The amount of blood required can be obtained by fingerprick or heelprick. If glassware is routinely cleaned with dichromate, it is advisable to keep separate that which is used for materials coming into contact with the enzyme and to use a detergent (such as haemosol) to avoid the excessive amount of rinsing required for adequate removal of the dichromate ions. A method is available which has proved suitable for use in a clinical laboratory [*20*]. A similar method apparently equally suitable has been published by Fleisher and Pope [*27*].

SUMMARY AND CONCLUSIONS

1. The literature relating erythrocyte cholinesterase titers (RBC ChE) to erythropoietic function is reviewed.

Normal reticulocytes and young red blood cells are far richer in this enzyme than are older red blood cells. Reticulocyte responses are accompanied by large increases in RBC ChE, and the high titers persist after the reticulocytosis is past. A relatively small preponderance of young cells results in RBC ChE levels distinctly above normal, and this can occur in the absence of a significant increase in reticulocytes. The cumulative effect makes it possible to detect small or gradual responses not reflected in the reticulocyte count.

2. Data on various hematologic disorders are reported and applications of RBC ChE to diagnosis and to the evaluation of erythropoietic function are illustrated and discussed.

In hereditary spherocytosis high titers were found consistently and persisted for years after clinically curative splenectomy. A significantly high RBC ChE was found in two of five apparently normal relatives of patients with the disease, suggesting that they carried the trait in a latent form not detectable by methods in common use.

Thalassemia major in an adult with evidence of chaotic acceleration of erythrocyte production and destruction was accompanied by exceedingly high RBC ChE. In three cases of thalassemia minor a normal RBC ChE was found even when the anemia was fairly severe. In sickle cell anemia our results confirm the finding of high RBC ChE reported by others.

In acquired hemolytic disease not associated with known underlying disorders the RBC ChE was high when the disease was active, normal in remission. When hemolytic anemia appeared in the course of malignant hematologic disease, the results were extremely variable and correlated with other evidence as to the erythropoietic status of the patient.

In patients with leukemia and multiple myeloma, frequent determinations of RBC ChE

revealed trends which were of prognostic significance. Upward trends from low to mid-normal or high normal occurred suddenly and preceded or accompanied the earliest other signs of remission, while downward trends were early signs of exacerbation and were a frequent although not invariable feature of the terminal period.

In a case of erythroblastosis a sudden rise in RBC ChE was followed by clinical improvement. In microcytic, hypochromic anemias the small red cells usually exhibit as much enzymic activity *per cell* as do normal red cells. The result is high activity *per unit volume* of cells which can be misleading if a correction for cell size is not made. Rarely in severe cases activity per cell has been below normal.

Observations in miscellaneous conditions are summarized. These include thrombocytopenia and purpura, hypoplastic anemia, anemia of chronic renal insufficiency, carcinomatosis, nonhemolytic icterus and minor blood loss.

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Seminar on Mycotic Infections

Current Concepts of Diagnostic Serology and Skin Hypersensitivity in the Mycoses*

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INTRODUCTION of biologically active material into a host will often induce certain responses dependent on the particular invasive organism. These responses may be employed in the identification of the microbiologic agent, whether it be virus, bacterium or fungus. Some of these responses are associated with changes in the circulating serum, changes which can be determined by one or more of a variety of procedures and technics for measuring circulating antibody. The serologic methods used with fungi are fundamentally similar to those used for the study of many infectious diseases. They are different in that the source of antigen is usually a more highly complex and differentiated organism.

The purpose of this article is to describe (1) the detection of antibodies to fungus pathogens by means of serologic tests, and (2) the appearance of hypersensitivity in animals or human subjects exposed to a given mycotic agent. The identification and evaluation of circulating antibodies, as well as the degree of hypersensitivity, may play an important part in diagnosis, prognosis and management. The mycotic diseases to be considered are histoplasmosis, cryptococcosis, coccidioidomycosis, candidiasis (moniliasis), North American blastomycosis and actinomycosis.

Certain characteristics of the serologic tests associated with pathogenic fungi are noteworthy. Fungi may be considered to be weak antigens, because concentrations of circulating antibody are usually low. In some tests detection of antibody in any titer may be indicative of a fungus infection. Thus the mechanics of the test may be of great importance, and the interpretation difficult and critical. Much work in coccidioidomycosis and histoplasmosis has been done toward standardization of methods and antigens, but many difficulties still remain. Why the pathogenic fungi elicit such a poor host response has not been established. Possibly the large size of whole organisms, such as Blastomyces dermatitidis and Cryptococcus neoformans, is not conducive to cytolysis or digestion by the host, with concomitant reduction of antigen release. Possibly the antigenic fractions are bound tightly within the cell wall and are released slowly into the host tissues. These characteristics, plus a paucity of slow growing organisms, might induce a condition of low concentration of circulating antibodies characteristic of some of the systemic mycoses.

Antigens presently used in serologic tests for the mycoses may show varying degrees of cross reaction. Much caution must therefore be exercised in the interpretation of results, and tests with antigens derived from related, cross-reacting species should be conducted before a definite diagnosis is made. This is especially true in the histoplasmosis-blastomycosis-coccidioidomycosis group, but applies also to all tests discussed in this paper. Fortunately, with the exception of Actinomyces bovis and its cross reactions in tuberculous animals, the fungus antigens under discussion do not generally react with antiserums to bacteria, rickettsiae or viruses. Thus a positive serologic test indicates a mycotic agent as the cause of infection.

Recently, endotoxins have been found to exist in many of the pathogenic fungi [*1*]. However, the development of antitoxins and the use of toxin-antitoxin reactions in diagnosis have not been reported.

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HISTOPLASMOSIS

At least three serologic tests and a skin test have proved of value in the diagnosis and prognosis of histoplasmosis: qualitative precipitation tests [*2,3*], quantitative agglutination tests [*4–7*], and a quantitative complement fixation test [*8–12*]. The precipitation and agglutination tests seem most helpful in newly acquired infections; for infections of long duration the quantitative complement fixation test has been of greatest assistance.

Technic of Tests. Two antigens derived from Histoplasma capsulatum have been used in precipitation tests: a filtrate from mycelial growth in an asparagine-glucose medium [*3*], or a filtrate from the growth of the yeast phase in an amino acid medium. The mycelial filtrate required several months to prepare, while the yeast phase filtrate was prepared from fourteen-day cultures. The mycelial filtrate tended to precipitate spontaneously when stored at 4°C. and therefore required filtration at regular intervals, whereas the yeast phase filtrate was stable. The writer has been using a precipitating antigen prepared in the following manner: Cells of the yeast phase are grown, under constant rotation, for fourteen days at 37°C. in 500 ml. flasks containing 100 ml. amino acid liquid medium [*13*]. The yeast cells, after being killed by exposure to 0.5 per cent formalin for seventy-two hours at room temperature, are separated from the broth by centrifugation and filtration. The broth is dialyzed against 0.85 per cent sodium chloride to remove the formalin, and then concentrated tenfold by evaporation. The solution is further dialyzed against physiologic saline solution and stored at 4°C. with 1:10,000 merthiolate as preservative.

Precipitin Test. The precipitin test is carried out by drawing equal volumes of antiserum and antigen into 3 inch lengths of capillary tubing. A control of uninoculated medium is included in each test. The mixtures are incubated at 37°C. for two hours, and refrigerated at 4°C. for thirty-six hours before final results are recorded. Three antigen dilutions are now used, undiluted, 1:10, and 1:100. This is a qualitative test, and several antigen dilutions are used only to overcome the possible effects of prozoning due to antigen excess. It is not a ring test, since definite precipitation takes place. Serums that are contaminated, hemolyzed or lipemic are not suitable for this test.

A precipitin reaction in agar, adapted from the methods of Ouchterlony [*14*], has recently been used to detect anti-H capsulatum antibodies [*15*]. In this test, about 25 ml. melted agar (1 per cent agar in 0.85 per cent sodium chloride plus a preservative) is poured into a Petri dish and allowed to solidify. Wells in the agar are punched out with a ¼ inch cork borer around a single central well. The central well is filled with 0.2 ml. histoplasmin, and each of the surrounding wells then receives an equivalent amount of unknown undiluted serums. Plates are incubated at room temperature and examined after forty-eight or seventy-two hours for zones of precipitation.

Complement Fixation Test. The quantitative complement fixation test can be conducted according to the method in use in any laboratory. Variations in methods, however, may affect the titer quite strikingly. In order, therefore, to attach the correct prognostic or diagnostic significance to a titer, suitable positive and negative serum controls should be included. In this laboratory the primary incubation period is one hour at 37°C. The system includes 0.2 ml. serum dilution, 0.2 ml. optimum antigen concentration, and 0.2 ml. of 2 units of complement. The hemolytic system of 0.2 ml. hemolysin containing 2 units and 0.2 ml. of a 2 per cent suspension of sheep erythrocytes is then added, and the complete mixture is incubated for another hour. The final reading is made after overnight refrigeration. Separate serum samples drawn at intervals of several weeks should be tested simultaneously so that rises or declines in titer may be detected and interpreted with greater accuracy.

Whole killed yeast phase cells [*8*] have been used extensively in the diagnostic complement fixation test. Two other antigens which have been employed are histoplasmin [*9*] and ground yeast phase cells [*10*]. Which antigen is best adapted for the diagnostic complement fixation test is still somewhat in doubt. For about ten years I used killed whole yeast phase cells as antigen and found it dependable for diagnosis [*3,8,16,17*]. Since some antiserums may react with yeast phase cells and not with histoplasmin, or vice versa, the recommendation has been made that both killed whole cells and histoplasmin be used as antigens in simultaneous tests [*12,18*]. Crude antigens should be used in preference to purified components [*10,19–22*] since it is not known which antigenic components of the cell produce the diagnostic humoral

antibodies. Also, no evidence has been presented to indicate that a single antigenic component is primarily responsible for the antibody response. Thus a crude "antigenic battery" may be more useful in such a test. The complement fixation test can be quantitated and used to study advance or regression of the infection.

Agglutination Test. A variety of technics has been used to demonstrate agglutinins against H. capsulatum antigens. Obviously, since cells of the yeast phase can be grown readily in large numbers in a variety of media, one of the simplest methods involves direct agglutination of killed whole cells of the yeast phase by immune serum [*23*]. Only recently, however, has such a test been described [*7*]. The cells were grown on brain-heart infusion agar for four days at 37°c., and inactivated by heat (65°c. for two hours), 0.5 per cent formalin or 0.5 per cent phenol. In a tube agglutination test, dilutions of antiserums were mixed with a stock antigen suspension, the tubes centrifuged immediately for seven minutes at 2,400 r.p.m., and the results observed. Unfortunately, direct agglutination of yeast phase cells has not been demonstrated with human antiserums but only with serums from hyperimmune or experimentally infected rabbits. Whether serums from proved or suspected cases of histoplasmosis in man can readily agglutinate yeast phase cells has not been established.

Other agglutination technics have been used. One test involves adsorption of histoplasmin onto sheep erythrocytes, and subsequent titration of rabbit antiserums for their hemagglutinating powers [*6*]. Another involves adsorption of histoplasmin onto either collodion [*4*] or latex [*5*] particles, and subsequent study of the agglutination of these particles in the presence of immune serums. In the histoplasmin-latex agglutination test, which provides more uniform suspensions than the collodion method, 0.1 ml. of a stock polystyrene-latex suspension and 0.5 ml. of an optimal dilution of histoplasmin were added to 9.4 ml. of physiologic saline solution and the resultant suspension washed. Dilutions of serum were mixed with the antigen, maintained at room temperature for two hours, and centrifuged at 2,000 r.p.m. for five minutes before final reading.

Meaning and Significance of Serologic Tests. In mild cases of histoplasmosis the only immunologic indication of a disease process may be conversion of the patient's skin test from negative to positive. This development of delayed hypersensitivity to histoplasmin, which is a filtrate from mycelial growth in an asparagine-glucose medium, is believed to develop about two to three weeks after the infectious process has begun. If the disease becomes slightly more severe but still remains essentially mild, precipitating antibodies may be detected in the patient's serum. These antibodies may persist for only a few weeks or months after infection, and their presence may be determined by precipitin or agglutination tests. Complement fixing antibodies may be completely lacking in such cases or may occur in moderately high titers, and great difficulty may be encountered in isolation of the etiologic agent by culture.

Complement fixing antibodies usually occur in patients with severe, chronic or disseminated disease. These complement fixing antibodies may first appear after the presence of precipitating antibodies has been well established. Complement fixing antibodies may persist longer, and certainly may behave independently of the latter. In many cases complement fixing titers may rise while precipitating antibodies disappear. The complement fixation test may serve both prognostic and diagnostic purposes. When the complement fixing antibodies are transitory, the prognosis is usually good. In some cases of histoplasmosis of extended duration the complement fixing antibodies tend to persist for long periods. This may be due to the presence within lesions of antigen in the form of dead or dying H. capsulatum organisms. Such lesions may possibly provide foci for endogenous reinfection at a later date. One pair of brothers, for example, still showed complement fixing antibodies (titers of 1:8 to 1:16) ten years after the first positive complement fixation test had been recorded [*24*]. Patients with extensively disseminated or terminal disease may show very low titers or no complement fixing antibodies.

The serologic tests for histoplasmosis have proved useful in directing efforts toward cultural isolation of the etiologic fungus. Thus, in many cases, cultural proof of the disease has come as a direct result of the detection of anti-H. capsulatum antibodies. The tests are also of value in the differential diagnosis of pulmonary lesions, so that the proper therapeutic approach may promptly be made, especially when tuberculosis is suspected.

It is essential, however, to appreciate the limitations of the serologic tests. In complement

fixation tests, some antiserums react to a much greater degree with yeast phase cells as antigen than with histoplasmin, or vice versa. Thus, unless both types of antigen are employed simultaneously in suspected cases of histoplasmosis, an error may result. In one study [*12*], for example, of 146 serums with complement fixing titers of 1:32 or more, 57 serums reacted to a higher titer with whole yeast phase antigen, while 41 serums were much more highly reactive with histoplasmin as antigen. In only about one-third of the serums, 48 in number, did similar titers develop with both antigens.

The absolute level of serum antibody, as determined by a particular test, is not important. A comparison of a series of serums from a given subject or animal provides data of diagnostic and prognostic value. Also, the level of titers of "normal" serums is of great significance.

Care should be exercised in selection of the complement fixation antigen, and in the assay for the optimum dilution. Some isolates of H. capsulatum provide superior yeast phase antigen, others provide superior histoplasmin [*25*]. Also, complement fixation activity of the antigen may vary with the method of growth, particular lot harvested, or the culture medium used [*26,27*].

It is unfortunate that the mycotic antigens, primarily from H. capsulatum, B. dermatitidis and C. immitis, may cross react to marked degree since there is often great difficulty in the clinical differential diagnosis of these infections, especially during the early stages of the pulmonary type [*28–30*]. In the complement fixation test, some serums from patients with histoplasmosis may cross react with antigens of B. dermatitidis. Characteristically, in such cross-reacting, complement-fixing serums the homologous yeast phase antigen produces a significantly higher titer. Serums from patients with histoplasmosis rarely cross react with C. immitis antigen. When such cross-reactions do appear, much higher titers usually develop with the homologous antigen.

Serums from patients with proved blastomycosis may sometimes react in equal complement fixation titers with H. capsulatum antigen, but are negative with coccidioidin. Serums from patients with coccidioidomycosis may react with the heterologous antigens from H. capsulatum and B. dermatitidis, but usually to a lower titer. Histoplasmin may cross react more completely than the whole yeast phase cell in a complement fixation test. In precipitin and agglutination tests, cross-reactions with histoplasmin as antigen are more extensive, and positive reactions accordingly should be judged with greater care. In serologic tests with the yeast phase of H. capsulatum as antigen, cross-reactions do not occur with serums from patients with nonmycotic disease, or with serums from patients with mycotic diseases such as actinomycosis, dermatophytosis, sporotrichosis or cryptococcosis.

Complement fixation and precipitation or agglutination tests should be conducted, since the latter tests are sometimes positive in milder cases of histoplasmosis, in which complement-fixing antibody never develops. Which type of precipitation or agglutination test is used, whether precipitation, red blood cell agglutination, collodion agglutination or latex agglutination, will vary with the particular laboratory. I prefer a capillary precipitin test with filtrate from the yeast phase as antigen because of the simplicity of the test and the completeness of the antigen.

Much effort has been expended in the attempt to obtain a purified specific antigen from H. capsulatum [*19–22,31*]. However, the recommendation is made that such purified antigens not be used in routine diagnostic serology until more knowledge is obtained concerning the sources and functions of the different antigens and antibodies. Some evidence exists [*19,22*] that individuals respond to a variety of antigens as the infection progresses or regresses. It may therefore be inadvisable for diagnostic purposes to restrict the test to the use of a single pure component as antigen. Further knowledge of the antigen-antibody reactions in diagnosis may result from studies such as those carried out by the Ouchterlony method [*15*]. With histoplasmin as antigen, at least six distinct precipitin bands have been identified from human antiserums, two bands recurring in seven patients that had culturally proved histoplasmosis. Such tests may indicate which antigen or antigens might be most frequently effective in inducing antibodies of diagnostic significance in patients with clinical histoplasmosis.

The exact function, if any, of the humoral antibodies to H. capsulatum is still in doubt, except that these antibodies do not seem to be related to increased resistance of the host [*32*]. In man, for example, high antibody titers are frequently found in cases in which the fungus is disseminating.

Skin Hypersensitivity. In a review in 1945, reports of only seventy-one cases of histoplasmosis could be found in the literature [*33*]. Ten years later, in 1955, 30 million Americans were estimated to have or to have had the infection. The basis for this radical change lies in the use of the skin test antigen, histoplasmin, as a tool to determine the epidemiologic pattern of the disease, to find new cases, and to aid in the confirmation of suspected cases.

The possibility of a benign form of histoplasmosis was first suggested by Smith [*34*] who had found many non-specific reactions to coccidioidomycosis in subjects from eastern central United States. With the aid of "a filtrate of whole broth culture of Histoplasma capsulatum" [*35*], a survey of 181 children in Tennessee indicated 53 per cent to be histoplasmin positive and tuberculin negative. In this group pulmonary calcification was present in seventy-eight, of whom 87.2 per cent reacted to histoplasmin. Three times as many children reacted to histoplasmin as to tuberculin. Later reports [*36,37*] indicated that 97.4 per cent of residents in Tennessee with calcification reacted to histoplasmin whereas only 18.8 per cent reacted to tuberculin. In a study conducted simultaneously in 3,105 student nurses a striking correlation was established between pulmonary calcification and histoplasmin sensitivity [*38*]. Of the whole group 22.9 per cent reacted to histoplasmin, while 79.3 per cent of those with pulmonary calcification reacted to the antigen. In contrast, only 10.2 per cent of the entire group and only 21.4 per cent of those with calcification reacted to tuberculin. The highest percentage of positive reactions occurred in subjects from the regions of Tennessee, Kentucky, Missouri, Kansas, Iowa, Illinois, Indiana and Ohio. A high prevalence of histoplasmin sensitivity and of proved cases of histoplasmosis was also found in subjects from parts of Virginia and Maryland. The antigen employed was prepared by growing strains of H. capsulatum on a synthetic fluid medium made in the manner recommended for the preparation of coccidioidin [*34*].

Crude histoplasmin is still being used for skin testing. Standardization of the antigen has been attempted against histoplasmin H-15 [*39*], although in many surveys it is not clear whether or not dilutions are comparable to the original H-15. At optimum dilution the antigen produces the maximum number of homologous responses and the minimum of heterologous responses [*40*]. In man, 0.1 ml. of antigen diluted to correspond with 1:1000 dilution of the original H-15 is injected intradermally. The reaction is usually read at forty-eight hours, although some responses present at twenty-four hours have disappeared by forty-eight hours. In some cases reactions are not manifest until seventy-two hours after intradermal injection, or even later. The diameter of induration is measured, 5 mm. or more being considered indicative of past or present infection. Erythema in the absence of induration is not considered a positive reaction.

A positive skin test to histoplasmin has the same significance in histoplasmosis as a positive tuberculin test has with regard to tuberculosis [*41*], namely, past or present infection with H. capsulatum. A negative reaction does not necessarily rule out the disease, since in the terminal stage of disseminated histoplasmosis the skin test may become negative. Also, under such circumstances as severe illness, temporary reversion from a positive to a negative reaction may occur [*42*]. Some subjects who react to histoplasmin may occasionally react, usually to a lesser degree, to blastomycin or coccidioidin. This cross-reaction, however, should not deter the physician from use of the histoplasmin skin test as an aid in diagnosis or as an epidemiologic tool. Four antigens, namely, histoplasmin, coccidioidin, blastomycin and tuberculin, should be used simultaneously in the skin test. The disease associated with the antigen which produces the greatest response should be the one to which emphasis is given in further laboratory procedures.

Since histoplasmin is now used extensively as a skin-testing antigen, it is essential to know whether or not reactions in a given serologic test are altered by prior intracutaneous injection of this antigen. After only one skin test, skin-positive subjects do not yield antibodies demonstrable by collodion-agglutination or by a complement fixation technic with a soluble extract from the yeast phase as antigen. Repeated positive histoplasmin skin tests do not cause increases in antibody titers with whole cells of the yeast phase as antigen but induce a detectable booster response with histoplasmin or the supernate from ground cells of the yeast phase as complement fixing antigen. Caution should therefore be exercised in interpretating antibody titers after the patient has been subjected to repeated skin tests.

Some attempts have been made to purify an antigen for intracutaneous tests [*43–45*]. A

single antigenic component has been isolated from the yeast phase which is active in inducing delayed type reactions in sensitized guinea pigs and man [*31*]. Physicochemical analysis reveals the presence of a complex containing carbohydrate and protein moieties, which moves as a unit in ultracentrifugation and in agar gel (in the presence of antibody). A quantity as low as 1.0 μg induces induration as great as or greater than that induced by standard histoplasmin.

CRYPTOCOCCOSIS

The amount of immunologic work performed on this mycosis has been limited, possibly because of the comparative ease of differential diagnosis by morphologic methods, possibly because of the apparent variation in antigenicity of different strains. The large encapsulated cells of Cryptococcus neoformans usually produce almost no inflammatory reaction, and as a result little if any host response. This indication that the body seems to make no effort to resist the infection presents an unusual immunologic picture, since evidence is being gathered that large numbers of subjects are exposed to the organism because of its presence in soil and the environment [*46,47*].

The statement has been made that the fungus is a poor antigen because the capsule limits contact of the cell proteins or antigens with the host tissues. Accordingly, some difficulty would be anticipated and actually is encountered in the detection of specific antibodies. The fungus, however, may not be a poor antigen but may actually produce an excess of antigen from the polysaccharide capsule. The host would thus have an excess of antigen, and immune bodies would not have the opportunity to manifest themselves or to function. Antibodies may be lacking because normal subjects may resist the fungus so completely that no striking immune response or lesion ever results. Only those subjects who are lacking or are abnormal in some aspect of immunologic resistance acquire the infection. Pulmonary cryptococcosis may occur in many people in a form too mild to detect by cultural or serologic means. Analogies may be made with such mycotic diseases as histoplasmosis and coccidioidomycosis, in which skin tests have been of great importance in establishing the frequency of infection. Similar skin tests with antigens from C. neoformans have been carried out but there has been little success thus far in establishing the range or frequency of the infection.

Serology. The genus cryptococcus has been divided into four morphologic groups, each group being distinct serologically, with little if any cross agglutination or precipitation between groups [*48*]. Strains of group 3 formed one serologic species, C. neoformans, but this included virulent strains causing fatal infections in man or animals as well as avirulent strains found as harmless parasites in normal man [*49*].

High titered rabbit antiserums have been produced by series of injections of the whole cells [*29,50*]. By a comparison of cross-reactions between nineteen strains of C. neoformans, three basic serologic types, A, B and C, were described [*51*]. The capsule is believed to be concerned with type specificity, as judged by its reaction to antiserums. The capsular polysaccharide was purified and isolated, and with the aid of filter paper chromatography three monosaccharide units were identified, namely, xylose, mannose and galactose, as well as a uronic acid [*52–56*]. The somatic proteins of a heavily encapsulated strain and a lightly encapsulated strain were isolated and compared [*57*]. The over-all amino acid composition of the two proteins was similar, both having a high content of dicarboxylic amino acids and lacking methionine. C. neoformans was also shown to produce an extracellular starch under special growth conditions [*58,59*]. This substance, after purification, had the properties of an amylose, although it contained an estimated 4.6 per cent protein as an impurity [*60*].

Diagnostic procedures employing serum from patients as a source of antibody have not been too successful [*61–66*]. A different and more successful approach toward diagnosis has been tried, using spinal fluid, blood and urine from a patient with cryptococcosis as antigen rather than as antibody [*67*]. The fluids were cleared by centrifugation and a series of dilutions of the supernatant mixed with an equal volume (0.2 ml.) of a potent anti-cryptococcus rabbit serum; precipitation was observed. Readings were made after two hours at room temperature and again after three days in the refrigerator. Complement fixation tests were performed with similar mixtures of supernatant fluids and serum, 0.2 ml. of each, plus 2 units of complement. All fluids from the patient gave positive reactions. In the case of the urine, tests were also performed with solutions of alcohol-precipitable material pre-

pared by three precipitations with 2.5 volumes of alcohol. The high titer of antigen found in the blood was of particular interest, since no cells of C. neoformans were demonstrable in the serum sample either by culture or by microscopic examination. Absorption experiments with solutions of purified cryptococcus capsular polysaccharide removed the capacity of the antiserum to react with the patient's spinal fluid and serum. These results indicate that the immunologic reactions are specific and that the reactive substances represent capsular polysaccharides. The precipitation and complement fixation results were further verified by quellung-like reactions of the capsule [*68,69*].

These data lend evidence to the viewpoint that the patient with cryptococcosis may have an excess of antigen. The capsular polysaccharide of cryptococcus may be difficult for the animal body to metabolize and therefore may remain in the cells and tissues for long periods of time. Obviously, this type of serologic test needs further experimentation and study, since it would be novel in fungus serology to use body fluids as antigens. Such use of tissue-antigen, however, has been made previously in bacterial diseases such as tularemia, in which the Ascoli test is used as an aid in diagnosis. This is a thermoprecipitin test in which tissues such as spleen are boiled, filtered, and the resulting supernate used as an antigen against specific immune serum [*70*].

Precipitating antigens may be obtained by one of the following technics: (1) emulsification and subsequent filtration of the organism in distilled water [*66*], (2) extraction with formamide according to Fuller's method [*71*], (3) disintegration of the cells by alternate freezing and thawing, then treatment of supernate with sodium acetate and ethanol followed by extraction with chloroform to remove protein [*72*]. The capsular reaction previously mentioned is similar to Neufeld's test for pneumococcus [*68,73*]. Washed cells either from infected animals or from agar culture, after mixture with immune serum and a small amount of a stain, such as aqueous eosin or methylene blue, showed prominent capsules with a sharply outlined border. No true swelling of the capsule occurred, merely an increase in the optical density of the outer part of the capsular substance. That this capsular reaction was caused by specific antibodies was indicated by the fact that the capsular reactivity of the serums was related to their agglutinin and precipitin titers, and also by the fact that the capsular reactivity was removed from the serums by the soluble specific antigens.

C. neoformans is serologically related to other species [*29,74*]. Cross-reactions have been established with hyperimmune anti-C. neoformans serums and Candida albicans, Saccharomyces cerevisiae and trichophytin, but not with antigens from H. capsulatum, B. dermatitidis and C. immitis. Cross-reactions were also obtained with pneumococcus type 2 and gum tragacanth against some anticryptococcal serums.

Hypersensitivity. Relatively slight attention has been given to hypersensitivity in cryptococcosis. In one patient there was a marked local skin reaction to a subcutaneous injection of a boiled aqueous extract of a C. neoformans culture [*75*]. In another patient there was a delayed reaction, 2 cm. in diameter, to a filtrate from broth cultures which had previously been killed by heating at 60°C. for two hours. Three "control" subjects showed no reactions to the antigen [*76*]. A delayed response to cryptococcus antigen, 4.5 by 5 cm. in diameter, was recorded in a patient with cryptococcal osteomyelitis [*77*]. Generally, negative results appear both in human patients and in experimental animals [*63,78*]. Whether skin hypersensitivity can be used as an aid in epidemiologic surveys or in diagnosis is still in doubt.

COCCIDIOIDOMYCOSIS

Immunologic studies have been of utmost importance in supplying specific information on the distribution, diagnosis and prognosis of coccidioidomycosis in both its primary and disseminated forms. These studies were greatly facilitated by such circumstances as (1) the highly endemic nature of the disease, (2) the introduction of large numbers of susceptible persons into military bases within the endemic area during World War II, (3) the opportunity to conduct immunologic tests on many subjects with infections ranging from the asymptomatic to the fatal disseminated type, and (4) the fact that the antigen coccidioidin is relatively specific in its immunologic behavior.

Coccidioidin and Skin Hypersensitivity. Much work prior, during and shorly after the war years was conducted under the supervision of Dr. C. E. Smith and his co-workers [*79–82*]. In precipitin, complement fixation and skin tests,

the antigen used almost exclusively has been coccidioidin, the filtrate from the growth of the mycelial phase of C. immitis in an asparagine-dextrose medium. The skin test with coccidioidin is generally recognized to be of great value in the clinical study of patients in whom coccidioidal infection is suspected. A positive skin test indicates that the patient probably has or has had a clinical or subclinical coccidioidal infection.

A greatly improved coccidioidin was prepared by growing ten strains of the fungus in an asparagine medium similar to the Bureau of Animal Industry tuberculin medium [*34*]:

L-Asparagine	7.0
NH_4Cl	7.0
K_2HPO_4 c.p.	1.31
Sodium citrate c.p. ($Na_3C_6H_5O_7.5\frac{1}{2}\ H_2O$)	0.9
$MgSO_4$ U.S.P. ($MgSO_4.7H_2O$)	1.5
Ferric citrate U.S.P. VIII (scales)	0.3
Dextrose (of the grade "cerelose") U.S.P.X.	10.0
Glycerine c.p.	25.0
Water to make	1000 ml.

After one to two months, when potency is indicated by preliminary tests, the broth is filtered and aqueous merthiolate is added to a final concentration of 1:10,000. The preparations are stable even at room temperature. Autoclaving does not seriously deteriorate the coccidioidin as an antigen in precipitin tests, but it does destroy the antigenicity for complement fixation. Even under apparently standardized conditions of inoculation and growth, marked variation may occur in potency and in anti-complementary behavior of the coccidioidin. Concentration has not been found to be feasible. Thus the contents of each flask should be tested before it is pooled with other flasks.

Most coccidioidins for skin testing have been compared in man with the reactions to a known lot, such as lot 9 of Smith [*79*]. If a dilution of 1:100 is used in surveys for the incidence of coccidioidomycosis, a dilution of 1:10,000 will suffice in patients with erythema nodosum. In cases of localized disease, a dilution of 1:1,000 should detect all the active infections, although in patients with pulmonary cavities of long duration dilutions as low as 1:10 may have to be used. In disseminated cases, dilutions of 1:10 may produce no allergic response. Caution should be exercised in the use of coccidioidin in low dilutions, since non-specific reactions may occur and unnecessary discomfort to the patient may be produced. The skin reaction characteristic of coccidioidomycosis is of the delayed type and usually begins to appear in six to ten hours, with the peak at thirty-six to forty-eight hours. Readings should be made at twenty-four hours to detect rapid reactors and at forty-eight hours to pick up slow ones.

The most sensitive method for diagnosis of an infection by C. immitis is the change from a negative to a positive reaction to coccidioidin. The incubation period from the time of infection to the first appearance of clinical symptoms is usually from ten days to two weeks, skin sensitivity appearing from two days to three weeks after onset of symptoms. However, in a study at four air fields in the San Joaquin Valley [*83*], 60 per cent of 1,351 infections were symptomless and only 25 per cent were diagnosed on the basis of clinical manifestations. In about one-sixth (77 of 462) of the patients with clinical cases of non-progressive disease, a test with 1:100 coccidioidin was negative during the first week of illness [*79*]. As the disease persisted the number of subjects with a positive reaction, and the severity of the reaction, rapidly increased, until by the fourth week of illness all were positive.

A difference of opinion exists as to the duration of coccidioidin hypersensitivity. A decrease or reversal in cutaneous hypersensitivity to coccidioidin over a period of twelve months was reported among twenty-one patients following their arrival at an area of low incidence of infection [*84*]. A person with a persistently positive skin test possibly may be exposed to repeated reinfection with consequent resensitization. Possibly, also, a patient with a persistently positive skin test may harbor a walled-off focus of living fungi, while those who lose sensitivity do not possess such a focus. This view is supported by observations in experimental animals in which pulmonary coccidioidal lesions became fibrosed and calcified and from which viable organisms could be recovered for at least two and a half years after infection [*85*]. On the other hand, no decrease in coccidioidin sensitivity was found among seventy-eight subjects with positive reactions in an area of low incidence of infection [*79,83,86*].

Although sensitization with broth culture filtrates has been reported in guinea pigs [*87*], no such phenomenon is believed to occur in man. Guinea pigs have been sensitized with killed mycelial suspensions, but not with coccidioidin known to be potent [*79*]. Continued retesting of subjects with negative reactions to coccidioidin in 1:100 dilution failed to induce hypersensitivity.

In an early attempt at purification and identification of the active substance in coccidioidin [*88*], an active principle was precipitated from a five months' growth in synthetic medium by the addition of 2 volumes of alcohol. The white, water-soluble powder contained 3 per cent nitrogen and on hydrolysis produced 20 to 40 per cent reducing sugars. Another purified polysaccharide contained no protein, but had 3.23 per cent total nitrogen and 0.6 per cent amino acid nitrogen [*89*]. The purified polysaccharide contained 11 per cent galacturonic acid and 79 per cent reducing sugars, of which 70 per cent was glucose and 30 per cent an unidentified sugar. As little as 0.01 μg of the purified substance produced positive skin reactions in sensitive persons. In the presence of immune human serums, positive precipitin reactions occurred, although there was no fixation of complement.

Coccidioidin is usually considered a relatively specific antigen. The first report of lack of specificity involved the reaction of coccidioides-sensitized guinea pigs to tuberculin [*90*]. The specificity of coccidioidin was further questioned because of the large number of reactions both in the general population and in tuberculous patients [*91*]. Kessel [*92*], however, found no cross-reactions with tuberculin in coccidioides-sensitized guinea pigs. The evidence for absence of cross-reactions of coccidioidin in tuberculous subjects was reinforced [*93*], when (1) the age distribution of coccidioidin sensitivity was shown not to follow the pattern of tuberculin sensitivity, and (2) Indians in the endemic coccidioidal area of southern Arizona had an 84 per cent coccidioidin reactivity and only 37 per cent tuberculin reactivity, while those in the non-endemic areas of northern Arizona had a 15 per cent coccidioidin reactivity and an 84 per cent tuberculin reactivity. Cross-reactions with other mycoses, such as histoplasmosis and haplomycosis, do occur [*34,94*].

The skin test offers the best index of the prevalence of infection with C. immitis in a community. In North America, the disease has been shown to occur in the southern half of California (particularly the San Joaquin Valley), Arizona, New Mexico, northern Mexico, western Texas, southern Nevada and southwest Utah.

Serologic Tests. The qualitative precipitin test may be performed by one of several methods. In a tube test [*80*], undiluted serum is mixed with undiluted antigen, 1:10 antigen and a control of uninoculated medium. The serum and the antigen are each used in 0.2 ml. quantities, and incubated at 37°C. for periods up to five days. Only a definite button is considered positive. Most precipitates form in twenty-four to forty-eight hours, although some serums require several days. Smith, therefore, recommends a five-day holding period. In most cases only the undiluted serum is necessary to detect a positive reaction. However, since a small number of serums may show prozoning because of antigen excess, a 1:10 dilution of antigen is included. In a capillary test, equal quantities of antigen and antiserum are drawn into a 3 inch capillary tube, incubated for two hours at 37°C., and refrigerated for thirty-six hours at 4°C. [*3*].

The complement fixation test, as with other tests for fungus diseases, can be performed with the technic which a laboratory is accustomed to using. Variations in complement fixation technics do affect the titer, and titer has been shown to have significant prognostic application [*80*]. The method of complement fixation used to determine the serologic pattern of 39,500 serums [*81*] was briefly as follows: 0.25 ml. of serum or serum dilution, 0.25 ml. of antigen, and 2 units of complement in 0.5 ml. were incubated for two hours at 37°C. The hemolytic system consisted of 0.5 ml. of a 2 per cent suspension of sheep erythrocytes sensitized with 2 units of hemolysin. The second incubation consisted of one hour at 37°C. and overnight refrigeration.

In mild unapparent infections, the serologic tests are usually negative. The more severe and prolonged the infection, the greater the possibility that the reaction will be positive. Serologic tests proved positive in 92 per cent of 732 soldiers with primary infections of sufficient severity to require hospitalization. Tests were positive in only 7 per cent of those with newly acquired asymptomatic primary disease.

Skin sensitivity to 1:100 coccidioidin is present in approximately five-sixths of the patients during the first week of symptoms [*79*]. Precipitins, however, may be detected in the serums of only half of these patients, fixation of complement in one-tenth. The precipitins, which appear before the complement fixing antibodies (if the latter do appear), reach their maximum during the second or third week of symptoms. Precipitins start disappearing notably after the first month of illness in primary non-disseminated cases, and by the fourth or fifth month have almost completely disappeared. Precipitins presumably

indicate active disease, the tests becoming negative when the infection is quiescent.

Complement fixing antibodies never reach the titers of precipitins. Complement fixing titers usually reach their maximum in primary non-disseminated infections by the second month and then begin to decrease. Complement fixing antibodies may persist longer than and independently of precipitins. In almost half the serums from patients with primary disease in which both precipitins and complement fixing antibodies are present, an increase in complement fixation titer occurs as the precipitins decrease. After several years, most complement fixation titers are not diagnostically significant. Subjects with healed coccidioidal lesions and a positive skin test are considered resistant to reinfection.

Serologic tests may be of great importance in following the course of infection and in warning of a threatening dissemination. Decreasing complement fixing titers indicate a receding infection. Observations on patients with disseminated infections have shown that the more severe the coccidioidal infection, the higher the antiserum titer to which complement will be fixed. Sixty-one per cent of 709 patients with disseminated disease had serum titers in excess of 1:16. When they exceed this critical titer the possibility of dissemination becomes great. This critical titer may vary greatly according to the sensitivity of the complement fixation test carried out in a particular laboratory [*82*]. A standardized, positive control serum can be used to aid in evaluation of results.

Patients with pulmonary cavitation fall in the same category as those with current primary non-disseminating disease [*95*]. In a study of 107 patients with pulmonary cavitation from whose sputums C. immitis was cultured, almost 40 per cent had negative or equivocal complement fixation titers. Of 192 patients with coccidioidal pulmonary cavity and positive complement fixing antibodies, 53 per cent were found to have complete fixation in a 1:2 dilution of serum, and 94 per cent had titers of 1:8 or lower.

Coccidioidin as a serologic antigen is more specific than the antigens from B. dermatitidis or H. capsulatum. In the precipitation test the antigen seems somewhat less specific than in the complement fixation test. In serologic tests for the diagnosis of pulmonary disease, antigens from C. immitis, H. capsulatum and B. dermatitidis should be used. Characteristically, especially in coccidioidomycosis, antiserum fixes complement to a significantly higher titer in the presence of homologous antigen.

CANDIDIASIS (MONILIASIS)

Of the species of Candida, usually only Candida albicans has pathogenic properties. To be helpful to the clinician, serologic reactions, therefore, must distinguish between C. albicans and the saprophytic species. However, close relationships within the genus Candida have been demonstrated in the serums of hyperimmunized rabbits by agglutination and agglutinin absorption technics [*96–98*]. C. albicans seems more closely related to C. tropicalis and C. stellatoidea than to other species of the genus, such as C. krusei, C. parakrusei (parapsilosis) and C. pseudotropicalis. Anti-C. albicans serums also have been shown to have agglutinins to antigens from species of cryptococcus, endomyces, mycoderma, saccharomyces and willia. Absorption by cells of the heterologous genera leaves behind agglutinins effective for the identification of the candida group.

Agglutinins have been demonstrated by a slide technic [*99*] and a test tube technic [*100*]. Serums from apparently normal people have agglutinated cells of C. albicans, possibly because of the presence of the organism in the mouth, intestine or vagina. For example, of serums from 1,150 normal people, 259 or 22.5 per cent agglutinated C. albicans although only thirty-five of these serums had titers of 1:160 or higher [*101*]. Agglutinins were present in the serums of 30 per cent of 533 females and 16 per cent of 617 males. A high titer correlated with the presence of the organism in the mouth or throat. In another study [*99*], 89 per cent of serums from 114 normal persons agglutinated C. albicans cells, mostly in low titer. These antibodies were believed to represent normal serum constituents.

Precipitins have been studied in the immunologic reactions of species of candida. Antigens were usually prepared by extraction of yeast cells with various solvents, although some experiments have been conducted with culture filtrates as antigens. These tests have been of little diagnostic use, although they have provided information on antigenic relationships within the genus candida.

Precipitinogens, which reacted with serums from a patient with bronchomoniliasis [*102*], have been demonstrated in alkaline extracts

of the organism. Treatment of this alkaline extract with alcohol produced a more concentrated antigen. A purified carbohydrate may be obtained from an alkaline extract of the yeast cells by precipitation with Fehling's solution, resolution of the precipitate with dilute hydrochloric acid and further purification by repeated precipitation with alcohol [*103,104*]. This carbohydrate is extremely active in the presence of hyperimmune rabbit serum, but shows cross-reactions at low dilutions with serums from animals immunized with C. parakrusei [*109*]. The supernatant fluid from autoclaved yeast cells [*98,106*] and the unheated alkaline extract of washed cells [*102*] apparently react well as a complement fixing antigen.

Skin Hypersensitivity. Hypersensitivity is an important feature in patients harboring C. albicans. Work has been reported (1) on skin tests with oidiomycin and (2) on the moniliid reaction. One of the first papers on allergic reactions to candida [*110*] showed that patients with intertriginous lesions from which "yeasts" were cultivated reacted to intradermal inoculation of an extract from the yeast. Successful treatment of these skin lesions resulted in a decrease and eventual disappearance of the skin hypersensitivity. The reaction to oidiomycin (an aqueous extract of C. albicans) is considered to be of the tuberculin or trichophytin type, in that the reaction is not in itself diagnostic but indicates an association with the organism sometime in the past. Conversely, a negative reaction would indicate insufficient contact with the organism to produce hypersensitivity or, in contrast, the presence of the organism in excess in the tissues so that anergy results. In one series of patients [*111*], 57 per cent of those with candida infection gave a skin reaction to oidiomycin, 43 per cent had no distinct reaction. In 45 per cent of the control subjects the reaction was positive, in 46 per cent negative, and in 9 per cent the results were equivocal. These experiments suggest the possibility that skin hypersensitivity may result from exposure to the organism without the development of clinical manifestations or that the patient's skin may become sensitized by continued presence of the yeast in such areas as the intestinal tract.

Certain trends from experimental studies on allergic reactions to the candida species may be summarized [*112*]. Spontaneous infection with species of candida may give rise to an allergic state, manifested by the development of generalized eruptions ("ids") and by reactions occurring after intradermal testing. Since the moniliids may be severe and chronic, such an allergic state may be very distressing. Some evidence, both clinical and experimental, suggests that sensitization may in some cases be a prerequisite for infection. Clinical observations indicate that sensitization is broadly specific in that cross-reactions with trichophytin and sporotrichin are lacking but are common within the candida group. The ability of a strain to sensitize is correlated with its potential virulence. Thus yeasts of human origin induce greater hypersensitivity in rabbits than do saprophytic yeasts, and species of candida pathogenic for rabbits induce more hypersensitivity than species of candida not pathogenic for this species.

NORTH AMERICAN BLASTOMYCOSIS

North American blastomycosis is readily confused with other conditions. Diagnosis based on clinical history and physical findings is only presumptive, and definitive diagnosis ultimately depends on the demonstration and identification of the etiologic agent, B. dermatitidis. An immunologic test, therefore, which can direct efforts toward a search for the correct causative organism is of great help.

Serologic Tests. One serologic test, complement fixation, has been of definite assistance as a diagnostic aid in North American blastomycosis. Again, any standardized complement fixation test may be used to measure the antibodies. As early as 1925, serums from patients with blastomycosis were shown to fix complement in the presence of mycelial suspensions as antigens [*113,114*]. However, the yeast phase has proved to be a more potent antigen and hence has been more widely used [*115–119*]. The yeast phase cell may be grown at 37°C. for four to seven days on a solid medium such as hemoglobin or blood agar, or in a liquid medium [*120*]. The organisms are killed by heat at 60°C. for four hours or by overnight exposure at 37°C. to 0.5 per cent formalin. The cell suspension is then washed, subcultured to insure sterility, and a preservative, such as 1:10,000 merthiolate, added.

One of the difficulties in performing the test is the preparation and titration of a suitable antigen. The yeast phase, regardless of how it is grown and how the cells are killed, may be found to have a low binding power, high anticomplementary properties, or both. The antigen

may also become more and more anticomplementary on standing, a property which may necessitate frequent preparation of freshly killed cells. Strains of B. dermatitidis seem closely related, and any strain therefore may be used as an antigen in the complement fixation test [*117*]. The broth filtrate, blastomycin, usually is not used as a complement fixing antigen in studies of blastomycosis, but has been used as a control antigen in serologic studies in histoplasmosis.

In early, mild or localized cutaneous infections, the complement fixation titer may either be lacking or extremely low. The titer increases with advancement of the disease and may remain high through the terminal stages. In a report of seventeen cases [*119*], serums from nine patients fixed complement in dilutions as high as 1:32. The titer of complement-fixing antibodies in a patient's serum reflects, within limits, the degree of involvement of the tissues and a fall in titer suggests clinical improvement. With recovery of the patient, antibodies gradually disappear from the circulation. High complement fixation titers portend a poor prognosis. At the same time, patients in the terminal stage of the disease may have a large number of fungus cells in their tissues and show no circulating antibodies by the complement fixation test.

A positive serologic test with a single fungus antigen may be misleading. Like serums from patients suspected of having histoplasmosis, serums that give positive titers with B. dermatitidis antigen should also be tested with heterologous coccidioidin and H. capsulatum antigens.

Attempts have been made to purify the antigens [*121,122*]. A soluble antigen prepared by sonic vibration of washed yeast phase cells was used to test for complement fixing antibodies in 165 serums, including 69 samples from patients with proved cases of blastomycosis and 15 from patients with a clinical or suspected diagnosis of other fungus infections. The antigen produced positive results in 71 per cent of the cases of proved blastomycosis, a percentage which corresponded with that produced by the whole yeast phase. The soluble antigen had the distinct advantage of ease of storage and of handling in the serologic test. A soluble carbohydrate-rich substance from the yeast phase fixed complement in the presence of serums from highly immunized rabbits but not in the presence of serums from human patients with blastomycosis. On the other hand, a soluble protein-containing substance obtained from washed cells of the yeast phase disrupted by sonic oscillation fixed complement in the presence of both types of sera.

Other tests have been used in making the diagnosis of blastomycosis, although with less frequency. For example, agglutination of the yeast phase cells by human serum has been reported [*123*], although in very low dilutions. A low agglutination titer in the serum of immunized rabbits was confirmed by quantitative antigen-antibody studies when 0.05 mg. of agglutinin nitrogen per ml. of serum was found [*29*]. This serum had a titer of 1:16 in the complement fixation test, using a suspension of yeast phase cells as antigen. Human serum also agglutinated fresh mycelial suspensions to a titer as high as 1:150 [*114*]. However, difficulty is frequently encountered in agglutination, apparently because of the large size of the yeast cell or the irregular particle in the mycelial suspension. The particles may agglutinate in the presence of normal serum or may fail to agglutinate in serums from patients with proved blastomycosis [*113,115*].

Precipitation tests have been carried out with soluble products from the mycelium or from the yeast phase [*122*]. Not enough information, however, is available for proper interpretation of this test. There seems to be little correlation between precipitin titers and complement fixing titers in serums from patients with proved blastomycosis. This may be because precipitin titers develop early in the disease and disappear before a rise in complement fixing antibodies occurs.

Skin Hypersensitivity. A characteristic feature of infection by B. dermatitidis is development of the hypersensitive state. This reaction is evidenced by a localized edematous response to the intradermal injection of either (1) a suspension of ground mycelial particles, (2) cells of the yeast phase, or (3) a culture filtrate. The response reaches its peak in twenty-four to forty-eight hours, and then rapidly fades. Although the three antigens cited seem to produce a similar type of reaction, and although any of the three may be used, the broth filtrate of the mycelium (blastomycin) is now used almost exclusively, probably because of (1) its proved stability over long periods of storage time, (2) its non-particulate nature, and (3) its potency.

During recent investigations on non-tuberculous pulmonary calcifications, skin testing with

blastomycin has been carried on extensively. The skin reaction is not specific, as indicated by cross-reactions in guinea pigs infected with H. capsulatum and Haplosporangium parvum and then tested with 1:100 dilution of blastomycin. Of 136 patients without demonstrable blastomycosis, 35 reacted to 0.1 ml. intradermal injection of 1:1,000 blastomycin [*124*]. In a study of 7,194 school children in Hamilton County, Ohio, an area in which proved histoplasmosis and blastomycosis occur, reactions to blastomycin occurred in 12 per cent and to histoplasmin in 33 per cent. Only one person who reacted to blastomycin failed to react also to histoplasmin. In addition, the reactions to blastomycin were of smaller size than the reactions to histoplasmin. Hence the blastomycin reactions were believed to be cross-reactions rather than specific reactions [*125*]. In an attempt to eliminate this non-specificity, a purified fraction was derived by ethanol precipitation from yeast phase culture supernate. This fraction has produced delayed skin reactions in rabbits infected with B. dermatitidis in amounts as little as 0.01 μg [*126*]. Its specificity and reactivity in human subjects, however, has not been recorded.

In a survey of forty cases of blastomycosis four patterns of immunologic reactions were described [*127*]. In the first group of ten patients, which included those with either recent pulmonary invasion or localized skin involvement, the skin test was positive and complement fixation negative. In the second group of ten patients, who showed more extensive infection, the skin test was positive and complement fixation positive, the serum titers ranging from 1:2 to 1:32, eight of ten having titers of 1:8 or lower. In the third group of ten patients, which included those with extensive generalized disease, the skin test was negative and complement fixation positive, with titers ranging from 1:4 to 1:64, eight of ten having titers of 1:16 or higher. In the last group, comprised of patients in either the terminal or incipient stages of the disease, both skin and complement fixation tests were negative.

ACTINOMYCOSIS

There is a paucity of information on the problem of serologic diagnosis in actinomycosis, a disease produced by the anaerobic organism, Actinomyces bovis. Because of the confusion in nomenclature, and the frequently incomplete descriptions of the organism, difficulty is encountered in distinguishing between serologic studies on A. bovis and those on pathogenic or saprophytic species of nocardia and streptomyces. Part of this confusion is a result of Bostroem's incorrect statement in 1891 that the organism is aerobic and a rather prevalent saprophyte [*128*]. For many years, therefore, few attempts were made to distinguish between actinomycoses produced by anaerobic species and those produced by aerobic species.

Antibodies to A. bovis have been demonstrated in patients with the disease. Agglutinins to the fungus have been demonstrated in the presence of homologous serum from infected human beings. Although trouble is occasionally encountered because of autoagglutination of the cells, successful use has been made [*129*] of an antigen prepared by drying the growth from a broth culture and grinding it in 0.4 per cent saline solution. High agglutinin titers were obtained in serums from heavily infected patients; the serums from people with localized infections, such as the cervicofacial type, failed to produce any agglutination.

Agglutination tests with nine rough and four smooth strains of A. bovis as antigens [*130*] revealed three serologic types in the rough forms and only one type in the smooth forms. With the exception of one strain, the two forms did not cross agglutinate readily. Fifteen isolates from human lesions fell into a homogeneous serologic group, distinct from five isolates of bovine origin, with no cross-reactions between the two groups [*131*]. Neither group showed any serologic relationships with four aerobic strains tested. In another study of twenty microaerophilic actinomycetes from human, bovine, equine and porcine sources, two basic serologic groups were demonstrated by agglutinin-adsorption technics. Each group contained isolates from three or four of the different sources, and therefore could not be separated serologically according to host [*132,133*]. With the use of an agglutinogen from sonically disintegrated cells in saline solution, and a precipitinogen contained in the supernatant fluid from these disintegrated cells, cross-reactions between A. bovis and streptomyces or norcardia were shown to be either slight or absent [*134*].

Antibodies have been identified in patients with various types of actinomycosis, including the cervicofacial type, either by complement fixation or by hemagglutination [*135–142*]. Antigens used in the complement fixation test

TABLE 1

IMMUNOLOGIC TESTS FOR THE DIAGNOSIS OF SIX MYCOTIC DISEASES

Disease	Agglutination	Test for Circulating Antibody		Skin Test
		Precipitation	Complement Fixation	
Histoplasmosis	+ (Latex-histoplasmin)	+ (Yeast phase filtrate or histoplasmin)	+ (Whole cells of yeast phase and histoplasmin)	+ (Histoplasmin)
Cryptococcosis	±	±	±	±
Coccidioidomycosis	−	+ (Coccidioidin)	+ (Coccidioidin)	+ (Coccidioidin)
Candidiasis	+ (Cells of yeast phase)	±	±	+ (Oidiomycin)
North American blastomycosis	−	±	+ (Whole cells of yeast phase)	+ (Blastomycin)
Actinomycosis	±	±	±	±

NOTE: + = Tests of value in diagnosis in man.
± = Tests have been reported but are not sufficiently advanced for use in routine diagnosis in man.
− = No satisfactory test.
( ) = Recommended antigen for test.

included whole organisms, ground cells, culture filtrates, and an aqueous extract prepared by heating the organisms to 65°C. In general, the results with complement fixation agree with those with agglutination tests.

Soluble antigens for complement fixation and hemagglutination were prepared by disruption of mycelial fragments and subsequent isolation of a nucleoprotein fraction with 0.1 N sodium hydroxide or of a polysaccharide fraction with 1 N sodium acetate [*142*]. These antigens are not specific; they may cross react with serums from tuberculous patients and from patients with streptococcal infections. However, the titers with the homologous organisms are much higher. Some serums from actinomycotic patients also react with tuberculin-coated red cells.

A. bovis may be readily isolated from tonsils or carious teeth. Whether this organism can induce detectable antibodies under such conditions has not been definitely established. On the basis of our present knowledge, therefore, little assistance can be expected in diagnosis from a single serologic test. Much additional experimental work must be done, even though experimental infection of laboratory animals may be difficult to accomplish [*143*].

Skin hypersensitivity also has been studied as a diagnostic tool in actinomycosis. As early as 1903, guinea pigs immunized with a strain of A. bovis were shown to be hypersensitive to tuberculin [*144*]. Later, two patients with actinomycoses were described as showing a distinct cutaneous reaction to a culture filtrate of A. bovis [*145*]. The edema lasted for two to three days, the reaction site attaining a diameter of 5 to 8 cm. Normal subjects gave no reactions. However, in another group of experiments, thirteen normal subjects produced more marked skin reactions to an A. bovis antigen than five actinomycotic patients [*146*].

Single injections of A. bovis (without adjuvants) rarely produce infection in laboratory animals, although repeated inoculations do. The suggestion has therefore been made that allergic sensitization may be of fundamental importance in establishing infection [*146,147*].

SUMMARY

The accompanying table (Table 1) summarizes the tests and the antigens employed therein. Some of these tests, such as the complement fixation tests for histoplasmosis and coccidioidomycosis, have been employed extensively and are recommended for use as aids in diagnosis and

prognosis. Others, such as the skin tests for cryptococcosis and actinomycosis, require further study before their true value can be ascertained.

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Clinicopathologic Conference

Thrombotic Thrombocytopenic Purpura

STENOGRAPHIC reports, edited by Lillian Recant, M.D. and Stanley Hartroft, M.D. of weekly clinicopathologic conferences held in the Barnes and Wohl Hospitals, are published in each issue of the Journal. These conferences are participated in jointly by members of the Departments of Internal Medicine, Preventive Medicine and Pathology of the Washington University School of Medicine and by Junior and Senior medical students.

A WHITE housewife, thirty-eight years of age, was admitted to the Barnes Hospital on December 16, 1958. She died approximately twenty hours later.

The patient was comatose at the time of admission so that historic data had to be obtained from the referring physician and accompanying relatives. She had always been in excellent health until October 1958, when she consulted a physician because of nervousness. For these symptoms she was given combid spansules (isopropamide, 5 mg.; and prochlorperazine, 10 mg.; in sustained release form); approximately eleven capsules were taken. A blood count in late October was said to have been normal. Two weeks before admission to the Barnes Hospital, petechial hemorrhages were first noted by the patient. At that time she also noted the onset of moderately severe generalized headaches which were treated with tablets which contained isobutyl-allyl-barbituric acid, acetophenetidin, aspirin, and caffeine, but with minimal relief. Despite the persistence of these symptoms, she did moderately well, remained at home, and was able to conduct her usual activities. Consciousness and mental clarity were apparently unimpaired, but petechiae persisted. She was admitted to a community hospital on December 14, 1948 for evaluation.

Examination at that time revealed the presence of petechiae over the lips, abdomen, and lower extremities and a palpable tender liver edge 6 cm. below the right costal margin. Laboratory data were as follows: red blood cells, 2,000,000 per cu. mm.; hemoglobin, 5.5 gm. per 100 ml.; white blood cells, 11,100 per cu. mm.; platelets, 80,000; hematocrit, 18 per cent; the differential count was interpreted as showing a moderate left shift; bleeding time, fifteen minutes; coagulation time, five minutes; prothrombin time, 80 per cent of normal. The Coombs' test was negative; osmotic fragility and serum bilirubin were within normal limits. Stool examination was positive for blood. Repeat blood counts were essentially unchanged except for an increase in the number of leukocytes to 22,050 per cu. mm.

Definite deterioration of the patient's mental status was noted by her husband shortly after hospitalization, although she remained conscious and oriented. A bone marrow aspiration was performed; the megakaryocytes were said to have been plentiful. During the following thirty-six hours the patient had periods of being irrational and aphasic; she complained of transient numbness of the side of her face and left hand, and became progressively obtunded. On December 16, she received a transfusion of 500 ml. of fresh blood, and shortly thereafter a persistent temperature elevation developed. Approximately five hours before admission to the Barnes Hospital she became comatose, was noted to have a tarry stool and vomited a small amount of blood. At that time therapy with cortisone (three doses of 25 mg. each) and broad spectrum antibiotics was begun. Her urinary output on the day of admission was reported to have been 100 ml.; examination of the urine revealed a specific gravity of 1.023, 4 plus proteinuria, 2 red blood cells and many coarsely granular casts per high power field. She was transported here for further evaluation. There was no history of arthralgias, pleurisy, allergic manifestations, jaundice, or of exposure to other medications or possible toxic agents.

On physical examination the patient was found to be a well developed, well nourished, white woman who was comatose, but responsive to painful stimuli. The vital signs were: pulse, 132 per minute and regular; blood pressure,

120/80 mm. Hg; respirations, 20 per minute; temperature, 39.2°C. per rectum. Numerous petechiae were noted over the anterior abdominal wall and upper and lower extremities; they were most marked over the distal lower extremities. There were no ecchymoses or purpuric manifestations. The head was normocephalic without abnormality to palpation, percussion or auscultation. The scleras were not jaundiced. The pupils were dilated but reactive to light. Examination of the optic fundi disclosed large bilateral flame-shaped retinal hemorrhages. There were no signs of papilledema. Crusted blood was present about the lips.

Examination of the upper respiratory tract revealed no abnormalities. The neck was supple and the trachea was in the midline. There were coarse rhonchi and scattered posterior basilar rales throughout both lung fields. No abnormalities of the heart were noted. The abdomen was flat and symmetrical. The hepatic edge was palpated 6 cm. below the right costal margin in the mid-clavicular line. No other organs or masses were felt. The peripheral pulses were normal. Neurologic examination revealed absent corneal reflexes and a hypoactive gag reflex. There was flaccidity of all extremities, and the deep tendon reflexes were symmetrically hypoactive. No pathologic toe signs were elicited.

Laboratory data revealed the following: red blood cells, 2,640,000 per cu. mm.; hemoglobin, 7.6 gm. per 100 ml.; reticulocytes, 40.6 per cent; white blood cells, 21,600 per cu. mm.; and platelets, 5,280 per cu. mm. The differential count was myelocytes, 2 per cent; metamyelocytes, 4 per cent; band forms, 13 per cent; segmented forms, 61 per cent; lymphocytes, 11 per cent; and monocytes, 9 per cent. There were 17 normoblasts per 100 white blood cells. Examination of the red blood cells revealed striking anisocytosis and poikilocytosis, marked polychromasia, and a moderate number of spherocytes. Many small, cellular clumps of bone marrow were obtained by sternal aspiration. Microscopic examination showed normal granulocytic elements, many megakaryocytes, and tremendous erythroid stimulation (340 nucleated red blood cells per 100 granulocytic cells).

Analysis of the urine showed a specific gravity of 1.020, reaction acid, and 4-plus proteinuria; microscopic examination of the centrifuged sediment revealed full field of red and white blood cells. The urine was negative for bile. Stool and cardiolipin examinations were not obtained. Blood chemistry determinations were: carbon dioxide combining capacity, 17.6 mEq. per L.; chloride, 101 mEq. per L.; nonprotein nitrogen, 72 mg. per 100 ml.; blood sugar, 151 mg. per 100 ml.; serum albumin, 4.5 gm.; globulin, 1.5 gm. per 100 ml.; alkaline phosphatase, 4.3 Bodansky units; serum calcium, 9.2 mg. per 100 ml.; and serum phosphorous, 4.1 mg. per 100 ml. One L.E. preparation was negative. An electrocardiogram was interpreted as showing sinus tachycardia and low voltage.

The patient's course was one of persistent rapid deterioration. She remained comatose, and had a sustained temperature elevation (38.5° to 40.8°C.). Fresh petechiae continued to appear. A lumbar puncture was performed with the following results: opening pressure, 190 mm. of water; the cerebrospinal fluid was clear and contained no cells; total protein was 35 mg.; chloride, 124 mEq. per L.; sugar, 103 mg. per 100 ml. The spinal fluid glutamic-oxaloacetic transaminase was 45 units. Culture of the fluid was negative. Vigorous therapeutic efforts consisting of fluid replacement, antibiotics, and large doses of steroids (a total of 1,000 mg. of hydrocortisone administered intravenously) were begun. In preparation for fibrinolytic enzyme therapy, further evaluation of her coagulation properties was made; the Ac globulin was found to be 50 per cent of normal but no other abnormalities were detected. Streptokinase therapy was instituted during the last few hours of the patient's life. Despite these therapeutic efforts, the patient remained comatose and died quietly.

CLINICAL DISCUSSION

DR. CARL V. MOORE: This patient had an acute illness. She was thirty-eight years of age, a housewife, entirely well as far as we know until October of last year at which time she consulted her physician because of nervousness. He found that she had a normal peripheral blood count. On December 1 she noted the onset of headaches and the development of petechiae, particularly on her lower extremities. Her headaches were not easily relieved by ordinary types of therapy. On December 14 she was hospitalized at a local hospital in Joplin, Missouri, and stayed there for the next two days. Shortly after admission her husband noticed a change in her mental status and that she was at times confused. Numbness developed of the left side of her face and left

hand. Following the performance of a marrow aspiration, she had an episode of aphasia. Because of the aphasia, the findings of a hyperplastic marrow, the anemia and thromboctyopenia, her physician suspected that she had thrombotic thrombocytopenia purpura. After a transfusion, her temperature rose. She became comatose and had one episode of hematemesis. She was severely oliguric. Her urine specific gravity was 1.023 with 4-plus proteinuria and coarsely granular casts. She was given cortisone, and transferred to Barnes Hospital. Massive doses of hydrocortisone were given intravenously but she died eighteen hours later without having regained consciousness. The diagnoses considered most seriously were: thrombotic thrombocytopenia purpura, disseminated lupus erythematosus with hemolytic anemia and thrombocytopenia, and acquired hemolytic anemia coexisting with idiopathic thrombocytopenic purpura with an intracranial hemorrhage. Dr. Reinhard, is it possible to decide among these three possibilities?

Dr. Edward H. Reinhard: This patient presented a clinical picture most characteristic of thrombotic thrombocytopenic purpura. The second possibility mentioned was disseminated lupus, with a coexisting thrombocytopenia. The rapid course of the disease however would be more in keeping with thrombotic thrombocytopenic purpura than with lupus, and in addition, we had one negative L.E. test. I do not believe, however, that we can state with any degree of certainty that this diagnosis has been eliminated. The third possibility mentioned was certainly a real one, namely that there was a coexistence of a severe hemolytic anemia and idiopathic thrombocytopenic purpura. These two conditions are known to coexist not infrequently. Any one of these three possibilities would have accounted for the clinical picture.

Dr. Moore: I gather that thrombotic thrombocytopenic purpura is your first diagnosis?

Dr. Reinhard: Yes.

Dr. Moore: Dr. Berg, one of the things that concerned those who saw this woman during her rapid terminal course was the etiology of her coma. Did she have hemorrhage in association with the thrombocytopenia or was the comatose state related to another process, as for instance thrombotic thrombocytopenic purpura? It was for this reason that the spinal fluid examination was obtained. Would you tell us whether or not you think the clear spinal fluid rules out any significant degree of intracranial hemorrhage?

Dr. Leonard Berg: A clear spinal fluid is found in 10 to 15 per cent of patients who at autopsy prove to have intracerebral hemorrhage. Certainly, the clear spinal fluid rules out subarachnoid hemorrhage, but a rather large hemorrhage may remain confined to the substance of the cerebral hemisphere. Against that possibility is the fact that focal neurological signs were minimal. Coma was the most striking terminal neurologic feature and was not accompanied by any obvious weakness on one side or signs of mid-brain compression by a swollen temporal lobe which might have contained an intracerebral hemorrhage. The total neurologic picture is very much against massive intracranial hemorrhage and of course the spinal fluid offers some evidence against it.

Dr. Moore: Even if the spinal fluid had contained blood, a diagnosis of thrombotic thrombocytopenic purpura would not have been ruled out because one can certainly have intracranial hemorrhage with that disease. While one can have fever, varying neurological manifestations, hemolytic anemia, and thrombocytopenia all pointing strongly in the direction of thrombotic thrombocytopenic purpura there is no definitive laboratory procedure that can be carried out to make the diagnosis final. One can sometimes find the characteristic thrombotic lesions in biopsy specimens of spleens, lymph nodes, skin, or liver, but there is understandable hesitation in obtaining biopsy specimens when the bleeding manifestations may be so severe.

Dr. Reinhard and Dr. Berg believe the disease to be thrombotic thrombocytopenic purpura. Does anyone care to challenge that diagnosis as the most likely possibility, or to offer any other suggestions? If not, I would like to direct the rest of our discussion to this disease and to talk first about its pathogenesis. Dr. Harrington, will you discuss the nature of the thrombotic lesions, and the nature of the changes in arteries particularly at the arteriolar-capillary junction?

Dr. William J. Harrington: The disease is confined to the area Dr. Moore has mentioned and originally was believed to be caused by platelet thrombi although histochemical studies were not conclusive. At the present time, through the use of the fluorescein antibody technic, it is now recognized that fibrin is a major component although there may be some platelets present and conceivably also some

degenerate red cells. We know only a few things about pathogenesis. First of all, patients with this disease have an extracellular corpuscular defect in that transfused red cells or platelets are rapidly destroyed. Dr. Brittingham and Dr. Chaplin have shown that there is no transferable plasma factor which accounts for the thrombocytopenia. It would appear probable, therefore, that there is some abnormality, not in the plasma, which leads to depletion of red cells and platelets focally in the smaller arteries. The most tempting concept is that the disorder is primarily one of the smaller arteries leading, for some unknown reason, to the focal fibrinous depositions. Often one may obtain a history of a shortly antecedent exposure to drugs or chemicals or biological material such as vaccines or antiserums. This had led some people to suspect that thrombotic thrombocytopenia may be related to the Schwartzman reaction.

DR. MOORE: May I go back to the thrombotic and the arterial lesions? Which do you think comes first?

DR. HARRINGTON: On the basis of pathological studies it has been postulated that the arterial lesions come first. This conclusion is based on the observation that in some small arteries one can find subintimal lesions which are presumed to be a prelude to the intravascular lesions.

DR. MOORE: Dr. Chaplin, Dr. Harrington has already mentioned the possibility that the anemia is caused by an extracorpuscular defect. Would you comment further? I know also that you obtained a faintly positive Coombs' test in this patient.

DR. HUGH CHAPLIN: The anemia has not been very well studied. The disease is relatively rare and antemortem diagnosis has only recently become common. It does indeed seem to be an extracorpuscular defect although I do not believe the definitive study of transfusing such a patient's cells into a normal recipient has been attempted. We recently had an opportunity to study a patient in whom this disease was diagnosed antemortem, and we did measure the red cell survivial time using the Cr^{51} method. In this instance the Cr^{51} half-life was six days, which is very much shortened.

DR. MOORE: Did you use the patient's own cells?

DR. CHAPLIN: Yes. In that patient we also carried out scintillation counting over the liver and spleen. While there was no evidence for splenic sequestration of red cells by increased counts over the spleen, at the time of splenectomy samples of spleen and a biopsy specimen of liver were compared for relative radioactivity; there was four times as much radioactivity in the splenic tissue per gram of weight as in the liver tissue. I am not absolutely sure how this should be interpreted, but it certainly suggests that the spleen may have played some role in the destruction of red cells since the splenectomy was performed after about three-fourths of the red cells had been destroyed.

Various studies for antibody against red cells have been carried out in this syndrome. Various types of warm and cold agglutinins, cold and warm incomplete antibodies and hemolysins active at normal and at acid pH have all been negative. Most of these patients have a negative reaction to a direct Coombs' test; however, in the particular patient being discussed today we find a weakly positive reaction to the Coombs' test which was entirely of the non-gamma globulin type. This result may reflect the greater sensitivity of the serum we use and it may be that a positive Coombs' test of this type will be found more frequently in the future. One other point, plasma hemoglobin measurements were carried out on this patient's plasma and the level obtained was 100 mg. per cent. Since the normal value by our method is less than half a milligram per cent, there was strong evidence of marked intravascular hemolysis. In the patient reported by Adelson,* the plasma hemoglobin value was 90 mg. per cent. I do not think we know the mechanism of the hemolytic anemia but I am beginning to think of it more in terms of an autoimmune type of reaction than I had formerly. I hope someone will comment on the possible relationship of marked hemolysis to the thrombotic process. Certainly the clot promoting factors that can be extracted from the red cell membrane are of great interest; in a patient with diffuse thrombotic lesions coexisting with brisk intravascular hemolysis, one is inclined to speculate that the two phenomena are related.

DR. MOORE: Dr. Brittingham, is the thrombocytopenia also caused by rapid destruction of platelets?

DR. THOMAS E. BRITTINGHAM: I know of two instances in which normal platelets were transfused into patients with this disease. On both occasions the transfused platelets disappeared

* ADELSON, E. and STEFANINI, M. Studies on platelets. IV. Demonstration and characterization of heterologous (Forssman) platelet antigen. *Blood*, 7: 700–709, 1952.

very rapidly. These observations provide suggestive evidence for rapid destruction.

DR. MOORE: Then how would you explain your own failure to develop thrombocytopenia when you took the plasma of a patient with thrombotic thrombocytopenia?*

DR. BRITTINGHAM: I do not know the answer to that. The possibility always exists that the dose of plasma was too small, but I think it is curious that the same quantity of plasma from an ordinary patient with idiopathic thrombocytopenic purpura and platelet agglutinins will produce thrombocytopenia in a normal recipient.

DR. MOORE: Dr. Berg, can one find enough lesions in the central nervous system to account for coma and the other neurological manifestations of thrombotic thrombocytopenic purpura?

DR. BERG: Although the brain may often look normal on gross inspection, widespread lesions of the small blood vessels in the brain are almost a uniform finding in autopsy examination of patients with this disease. The many small lesions in the small blood vessels may or may not lead to obvious microscopic destruction of surrounding tissue.

In the past ten or fifteen years, neurologists have come to regard consciousness as a phenomenon having a great deal to do with a particular portion of the upper brain stem called the reticular activating system. Relatively small lesions in this system may cause profound impairment of consciousness. Consequently, with the combination of microscopic infarcts, the few petechiae, and the recognized phenomenon that impairment of blood supply may interfere with function of an area of the nervous system such as the reticular activating system without actual death of tissue, one can ascribe the profound neurologic manifestations to the small blood vessel lesions that are seen.

DR. MOORE: Dr. King, can one explain the fever on the basis of changes in the central nervous system or on the hemolysis?

DR. M. KENTON KING: Fever of this type occurs in either of the disorders you have mentioned; in this particular patient it would be difficult to select one cause for the sustained fever in preference to another.

DR. MOORE: Dr. Bricker, will you discuss the pathogenesis of the renal changes. Even though our patient had hemoglobin in her plasma, she seemed to tolerate the transfusion she received without difficulty. Can you explain why she had oliguria?

DR. NEAL S. BRICKER: When the patient was admitted to Barnes Hospital she had definite chemical evidence of a moderately severe decrease in the number of functioning nephrons. This was evidenced by the azotemia and by the acidosis. However, her phosphate concentration was normal and so was her calcium. The latter are inconsistent with terminal chronic renal disease. Yet she was oliguric and in the natural history of chronic progressive renal disease, persisting oliguria occurs only during the terminal phases. For this reason, it would seem reasonable to postulate that she had an acute renal lesion which was diffuse, extensive and devastating. However, if the only renal insult occurred on the day of transfer to Barnes, then the rise in non-protein nitrogen and the fall in arterial pH would have occurred too rapidly even though she was catabolic from fever and was receiving steroids. Perhaps the most logical explanation therefore is that she had an underlying renal lesion which was chronic or subacute but that she also had superimposed an acute lesion, possibly on the day of admission. One can postulate that the chronic lesion was the same process that may have involved arterioles throughout her body, namely arteriolar occlusions with attendant small infarcts.

DR. MOORE: Let me see if I understand you. You think she had thrombotic lesions diffusely throughout the renal parenchyma, but then on the day of admission may have had an acute insult as from a transfusion reaction.

DR. BRICKER: I think that she may have had a chronic or subacute lesion, but that this will not explain all the findings. Therefore, there also must have been an acute process occurring perhaps on the day of admission. I am not at all sure that it was secondary to a transfusion reaction. One possibility is that she had a rapid acceleration of her thrombotic disease which involved virtually all the arterioles of the kidney and resulted in the almost complete loss apparent on admission. The second possibility, of course, is that she did have acute tubular necrosis secondary to the transfusion reaction. However, there are several disturbing features about the latter diagnosis. One is that her urine was concentrated on two occasions. Secondly, there was a high concentration of protein in the urine on two occasions. Even allowing for the

* BRITTINGHAM, T. E. and CHAPLIN, H., JR. Attempted passive transfer of thrombotic thrombocytopenic purpura. *Blood*, 12: 480, 1957.

effect of protein on the urine specific gravity, there is still a possibility that this was a concentrated urine, that is, that the osmolality of the urine was greater than that of the plasma. This is extremely unusual in acute tubular necrosis. The other two somewhat unusual features are the numerous red cells in her urine (although she was thrombocytopenic) and the numerous white cells. Thus, although we cannot exclude the possibility that she had acute tubular necrosis I think it is relatively unlikely. Finally, this disease has been associated at least in theory with the Schwartzman reaction and in the Schwartzman reaction one occasionally sees diffuse bilateral renal cortical necrosis.

DR. CHAPLIN: Her hemoglobin rose 2.2 gm. in response to the transfusion. If she had a reaction it was probably not of the hemolytic variety.

DR. MOORE: Dr. Eisen, so far we have been discussing the pathogenesis of specific manifestations. I would like to focus for just a few minutes on possible etiological mechanisms. Several people have already pointed out that there is a certain resemblance between thrombotic thrombocytopenic purpura and the Schwartzman reaction. How close is the resemblance?

DR. HERMAN EISEN: With respect to the lesions which occur in the small blood vessels in thrombotic thrombocytopenic purpura, there is a close resemblance to the corresponding lesions of the generalized Schwartzman reaction. In both situations, fibrinoid material stains strongly with fluorescent antifibrin antibodies. This sensitive reagent indicates that the eosinophilic occlusive material in the small blood vessels is, at least in part, fibrin or fibrinogen, in both the disease we are discussing in man and in the experimental generalized Schwartzman reaction. In both situations there occurs also thrombocytopenia and leukocytosis. Beyond that however there are big differences. The hemolytic anemia, which was so conspicuous in this patient, has no counterpart so far as I know in the experimental Schwartzman phenomenon.

DR. MOORE: Dr. Harrington earlier pointed out that some people believe arterial lesions came first in thrombotic thrombocytopenic purpura. Is that also true for the Schwartzman phenomenon?

DR. EISEN: I do not know. There is some question about whether or not the intramural lesion does not contain fibrin deposits also.

DR. MOORE: Dr. Lange, are you in agreement with the people who have implied that thrombotic thrombocytopenic purpura is an autoimmune disease of some kind?

DR. ROBERT D. LANGE: Most investigators seem to suspect that there is a hyperimmune mechanism associated with this disorder.

DR. MOORE: Before concluding, we should spend a few minutes talking about therapy because everyone has an extremely pessimistic point of view about the treatment of this condition. Most patients die rather quickly, Dr. Mohler, but there have been a few reported instances in which patients have done moderately well after splenectomy for a period of time. What place should splenectomy have in the treatment of thrombotic thrombocytopenic purpura?

DR. DAN MOHLER: In a disease such as this, with such a poor prognosis, splenectomy should be given further trial. The cases recorded in the literature are few in number. Two patients survived a year after splenectomy, and two other patients were still alive three years after splenectomy.

DR. MOORE: Burke and Hartmann have recently described two patients who had rather long remissions following massive steroid therapy.* There is some reason then to continue therapeutic trials with massive steroid therapy, splenectomy, or both.

The question of anticoagulant therapy for this patient was discussed when she first arrived in the hospital. We have talked about the fact that fibrin and fibrinogen have been identified in the thrombi. Dr. Chaplin has wondered whether the thrombi could be initiated by the intravascular destruction of red cells. Would anticoagulant therapy be rational Dr. Perkins?

DR. HERBERT A. PERKINS: The association of thromboses with massive hemolysis does seem to exist, for example, in paroxysmal nocturnal hemoglobinuria which has perhaps as massive intravascular hemolysis as any condition we see. In this particular condition anticoagulant therapy has been suggested for this very purpose. Similarly, in the hemorrhagic syndrome that can follow a massive intravascular transfusion reaction, the group at Johns Hopkins has recently reported studies suggesting that the coagulation factors which have disappeared and lead to the hemorrhagic syndrome have done so because of the infusion of thromboplastin-like material. Red cell hemolysates have been shown to have

* BURKE, H. S., JR. and HARTMANN, R. C. Thrombotic thrombocytopenic purpura. *Arch. Int. Med.*, 103: 105, 1959.

thromboplastic activity. However, in thrombotic thrombocytopenic purpura, the lesions are not typical of the thromboses found in these other conditions, and the primary condition may be in the blood vessels rather than in an increased tendency of the blood to clot. Since we have severe thrombocytopenia, damaged blood vessels, and local hemorrhages anyway, I would be extremely hesitant to try anticoagulant therapy of any sort.

DR. MOORE: Dr. Fletcher, Dr. Reinhard persuaded you, in his desperation, to treat this patient with the intravenous administration of streptokinase. Would you comment on this approach to the therapeutic problem?

DR. ANTHONY P. FLETCHER: I am glad that you mentioned Dr. Reinhard's therapeutic desperation because this represents the current status of our knowledge. However, there is some evidence by which opinion may be guided. Although from a histochemical viewpoint there is doubt as to the composition of the vessel thrombi, I think that considerable emphasis should be placed on the results of fluorescent antibody studies which suggest that the vessel thrombi contain a substantial proportion of fibrinogen. Dr. Eisen has mentioned that the vessel lesions may arise secondarily to a generalized Schwartzman reaction. Now there is some evidence that streptokinase therapy will either prevent or relieve vascular lesions occurring secondary to the generalized Schwartzman reaction.* Consequently we decided that streptokinase therapy, despite its obvious hazards, was justifiable. In this particular patient, a further consideration arose but since our period of study was short, we have little evidence contributing to its elucidation. For streptokinase therapy to be rational, vessel thrombi in addition to being composed of fibrin must also contain plasminogen. Our limited biochemical studies showed plasma plasminogen concentrations to be only 30 per cent of normal and accelerator globulin also to be depressed, evidence suggesting that the patient had previously experienced a burst of plasma fibrinolytic activity. The presence of a normal plasma fibrinogen concentration and the absence of antithrombin activity suggested that the release of fibrinolytic enzyme had occurred several hours previously. These findings would indicate that streptokinase therapy could conceivably have been impeded by lack of thrombus plasminogen. We only treated her with streptokinase for a short while, whilst her condition appeared terminal and I do not believe that the final outcome contributes to therapeutic evaluation.

DR. KING: Relative to the comments of Drs. Eisen and Perkins, I would just like to say that the administration of anticoagulants will regularly modify the localized Schwartzman phenomenon.

DR. MOORE: Are there any additional comments?

DR. REINHARD: I want to be sure that the impression was not created that splenectomy is of proved value in this disease. Dr. Mohler pointed out four cases in which the patients did seem to survive longer than usual and to get some temporary benefits following splenectomy. However, there are many other cases in which splenectomy has been performed with no benefit whatsoever. Furthermore, while this disease usually runs a rapidly fatal course, there are rare instances in which the course is chronic with or without splenectomy.

DR. CHAPLIN: The patient I referred to earlier got very much worse immediately after splenectomy. He became much more febrile and his course went downhill rapidly thereafter. That patient also received three times the amount of steroid therapy that was given to the patients recently reported. This was to no avail.

DR. MOORE: In concluding, we may have been overconfident in our analysis of this problem, but we do not anticipate any great surprises from the pathologists. I believe this patient had thrombotic thrombocytopenic purpura with all of its attendant manifestations, and doubt that there will be any evidence of a hemolytic transfusion reaction.

PATHOLOGIC DISCUSSION

DR. ARA CHALVARDJIAN: The gross anatomical findings were those of a generalized purpura that involved many of the organs to a greater or lesser degree. Cutaneous petechiae were observed, especially over the trunk and lower extremities. Crusted blood was present over the lips. The pleural, pericardial and abdominal cavities each contained about 100 ml. of straw-colored fluid. The tracheobronchial lymph nodes were hemorrhagic. The epicardial surface of the heart showed numerous petechiae and even

* CONDIE, R. M., HONG, C. Y. and GOOD, R. A. Reversal of the lesions of the generalized Schwartzman phenomenon by treatment of rabbits with streptokinase. *J. Lab. & Clin. Med.*, 50: 803, 1957.

FIG. 1. Gross photograph of the epicardial surface of the heart which contained numerous, almost confluent, petechiae.

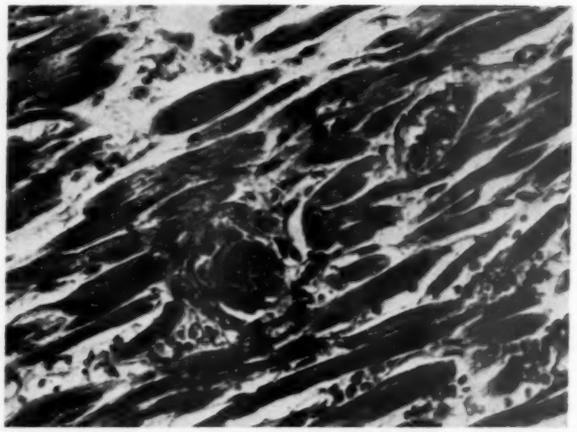

FIG. 2. Arterioles in the myocardium filled with thrombotic material. Note that the muscle fibers adjacent to arterioles appear compressed. Hematoxylin and eosin × 250.

larger, ecchymotic areas. (Fig. 1.) Prominent petechiae were also present throughout the myocardium, especially in the interventricular septum. The kidneys each weighed 150 gm. and a few petechiae were seen both in the cortex and in the medulla; otherwise they were within normal limits. The spleen weighed 320 gm. and was slightly enlarged, with prominence of the follicles. The liver extended 5 cm. below the costal margin and weighed 1,820 gm. but otherwise was within normal limits. The adrenals showed a few petechial hemorrhages. The stomach contained about 450 ml. of fresh blood, with some clots, and its mucosal surface was extremely congested, with shallow, recent ulcerated areas along the greater curvature. The femoral bone marrow was red, indicating hyperplasia of the hematopoietic tissue. The brain weighed 1320 gm. and many petechiae were present, widely scattered throughout the white and grey matter without any particular pattern of distribution.

DR. ROBERT M. O'NEAL: The gross lesions in this case provided a demonstration of disseminated purpuric lesions. Microscopically, the pertinent lesions were all those which we associate with classic thrombotic thrombocytopenic purpura. These lesions were most numerous in our microsections of the interventricular septum of the heart. (Fig. 2.) Here, the arterioles present between cardiac muscle fibers were often much larger than we would expect to see normally, presumably representing the aneurysms that have been demonstrated at

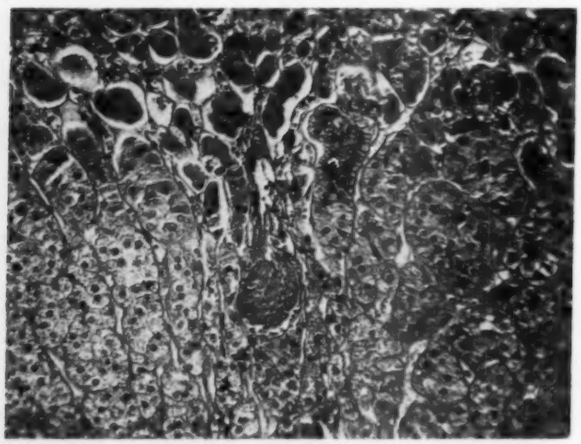

FIG. 3. Thrombotic material in a small vessel in the adrenal cortex. This thrombus appears layered, and may lie in a venule. Hematoxylin and eosin × 250.

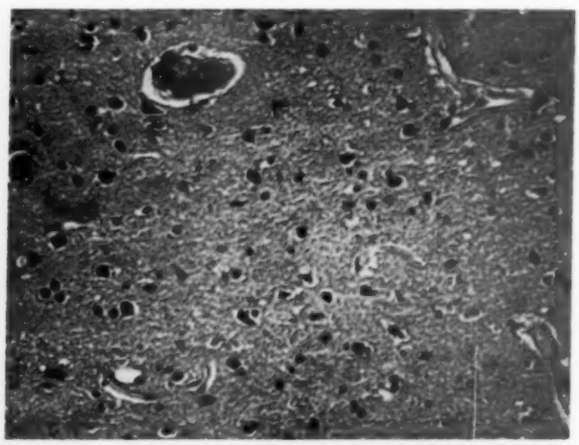

FIG. 4. No abnormality of the brain substance is seen here, but an arteriole is filled with amorphous granular material. Other areas contained petechiae. Hematoxylin and eosin × 250.

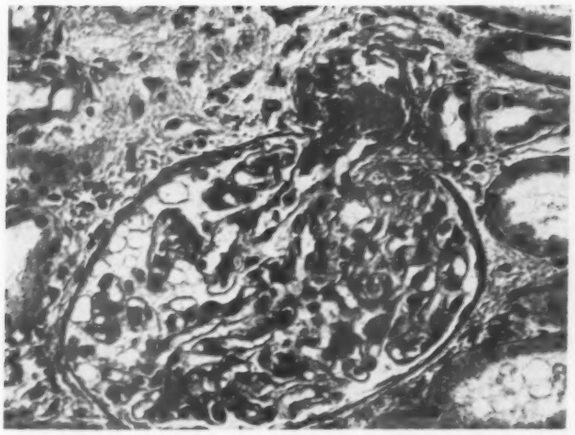

FIG. 5. One of the thrombotic masses lying in an afferent arteriole in the kidneys. The glomerulus does not appear to be altered. Hematoxylin and eosin × 250.

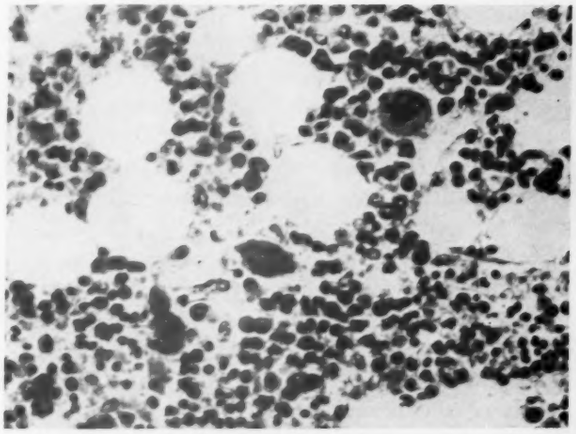

FIG. 6. Femoral bone marrow. The normally fatty marrow has been converted to hematopoietic marrow such as is usually found in vertebras and ribs. Megakaryocytes were prominent here as well as in the lungs. Hematoxylin and eosin × 250.

the arteriolar capillary junction by serial section and reconstruction by Dr. Orbison.* The walls of such arterioles were very thin but with routine hematoxylin and eosin stain specific lesions of the arteriolar wall were not seen. The thrombotic material filling arterioles was pink and granular, only occasionally appearing layered. Endothelial cells, or possibly some fibroblasts, were scattered through the thrombi. At high magnification endothelial cells could be seen forming a continuous covering over the homogeneous thrombotic material within arterioles.

With ordinary fibrin thrombi, the usual mixed thrombi we find at autopsy, the fibrinous component stains purple with phosphotungstic acid-hematoxylin, and in patients that we have suspected to have had a generalized Schwartzman reaction, the thrombotic material also was deep purple with PTAH staining. In the patient, as in others with this disease, the thrombotic material is uniformly negative with our usual fibrin stain. As Craig and Gitlin have shown *in vitro*, a low albumin concentration will affect PTAH staining of fibrin,† but of course in this patient the serum albumin concentration was far above the 1 gm. per cent critical level and still there was no staining with our usual fibrin stains. We simply do not know why this thrombotic material in thrombotic thrombocytopenic purpura, which apparently does contain fibrin, fails to take our usually reliable histologic stains for fibrin.

Silver staining has been used by others to demonstrate the difference between the thrombotic material in this disease and that in ordinary thrombi,* but we performed these and failed to demonstrate a difference to our satisfaction. Apparently these staining reactions are not quite as consistent as might be hoped.

Practically all organs were involved. In the liver, small arterioles containing thrombi often resembled bile ducts because of the extreme prominence of the proliferated endothelial cells. Arterioles in the spleen and adrenal were also involved. (Fig. 3.) Thrombi were frequent in small arterioles of the brain and, although no obvious areas of infarction were seen, there were numerous petechiae which might well have been superimposed on, and thus obscured, small infarcted areas. (Fig. 4.)

In the kidney thrombi were also widespread, often being found near glomeruli in the afferent arteriole just at its point of entrance. (Fig. 5.) There were no characteristic lesions of hypoxic nephrosis, and it seems likely that these thrombi, obstructing arterioles, were the cause of the patient's renal failure.

The femoral bone marrow was extremely hyperplastic, containing only the amount of fat that we might expect in the red bone marrow of the ribs and vertebras. (Fig. 6.) Megakaryocytes were very prominent throughout the marrow of

* ORBISON, J. L. Morphology of thrombotic thrombocytopenic purpura with demonstrations of aneurysms. *Am. J. Path.*, 28: 129, 1952.

† CRAIG, J. M. and GITLIN, D. The nature of the hyalin thrombi in thrombotic thrombocytopenic purpura. *Am. J. Path.*, 33: 251, 1957.

* FISHER, E. R. and CREED, D. L. Thrombotic thrombocytopenic purpura. *Am. J. Clin. Path.*, 25: 620, 1955.

all sites, appearing definitely increased in number. The number of nuclei in individual megakaryocytes appeared increased. Numerous megakaryocytes were also found in many of the pulmonary capillaries; many low power microscopic fields contained 3 or 4. They were also present in the glomerular capillaries. So we were able to demonstrate all the typical lesions of thrombotic thrombocytopenic purpura, as you suspected we would.

Final anatomical diagnoses: thrombotic thrombocytopenic purpura with involvement of all organs, generally; marked hyperplasia of all elements of the bone marrow, including femoral bone marrow; extramedullary hematopoiesis in the spleen; numerous megakaryocytes in capillaries of the lungs, spleen, and renal glomeruli; hemorrhage into the gastrointestinal tract, 250 cc. in stomach, tarry feces in colon; hydrothorax, 200 ml; hydropericardium, 100 ml.; and ascites, 125 ml.

DISCUSSION

DR. MOORE: Thank you. I do not think we should terminate this conference without several more comments. I am somewhat disturbed by the fact that these thrombi did not take a fibrin stain, and that no pathologist has ever been able to make them take this stain.

DR. EISEN: Gitlin and Craig have shown that for fibrin to give typical color reactions with phosphotungtic acid and similar reagents, it is necessary that the fibrin be formed in the presence of appreciable quantities of serum albumin. "Pure" fibrin does not stain with dyes as do some fibrin deposits in tissues. It is possible that the classic tissue staining reactions for fibrin are, in fact, not staining reactions for fibrin *per se* but for serum albumin which is concentrated on fibrin strands. One would expect that if a fibrin deposit formed in tissue in such a way as to be little, or not at all, contaminated by serum albumin the expected classic staining reactions for fibrin would be negative.

In respect to the specificity of the fluorescent antibody technic, this procedure can, of course, be no better than the purity of the antiserum which is rendered fluorescent. If the antiserum contains several species of antibodies each specific for a different antigen, as is usually the case, the fluorescent antiserum can stain more than one antigenic material. The closer one approaches to an antiserum of a single specificity the more closely one approaches a fluorescent reagent which is uniquely specific for a single antigen. In the case of antifibrin serum, I understand that appreciable serological reactivity is still present for fibrinogen. Hence the fluorescent antifibrin serum very likely does not distinguish between fibrinogen and fibrin. From the work of Gitlin and of Dixon and their associates I think it well established, however, that the occlusive vascular deposits in thrombotic thrombocytopenic purpura do contain fibrin or fibrinogen. The failure of these deposits to give the classic color reactions for fibrin is perhaps explained by the work of Craig and Gitlin on the importance of serum albumin in these staining reactions.

Case Reports

Giant Hypertrophy of the Gastric Mucosa, Hypoproteinemia and Edema (Menetrier's Disease)*

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New York, New York *Washington, D. C.*
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GIANT hypertrophy of the gastric mucosa was first described by Menetrier in 1888, who termed the lesion polyadénome en nappe. Synonyms include Menetrier's disease, giant hypertrophic gastritis, giant hypertrophy of the gastric rugae, polypoid swelling of the gastric mucous membrane, and adenopapillomatosis gastrica [*6*]. In most of the reported cases plasma protein determinations were not obtained; in those cases in which they were determined hypoproteinemia has frequently [*2*] but at times infrequently [*3*] been reported. Edema in the absence of other associated disease has been recorded in nine cases [*1–6,8–10*]. In four other instances severe anemia [*6,7*], renal disease [*2*], mitral disease and hepatosplenomegaly [*6*] were present; their contribution, if any, to the genesis of the edema is difficult to assess. The serum proteins were determined in only four subjects with edema and found to be depressed [*1,8–10*].

The tenth recorded case of edema and giant hypertrophy of the gastric mucosa, in the absence of associated disease, is herein reported.

CASE REPORT

D. S., a twenty-five year old Negro man, was admitted to the medical ward on July 26, 1957, with complaints of anorexia, vomiting, abdominal cramps and watery diarrhea of three months' duration. He complained of residual tightness in the stomach after meals, epigastric fullness, and refused to eat.

Physical examination revealed a blood pressure of 120/80 mm. Hg, a pulse rate of 94 per minute, and a temperature of 99°F. The head and neck appeared to be normal. The lungs were clear to percussion and auscultation. Examination of the heart revealed no thrills or murmurs. The abdomen was diffusely tender but there were no masses, and the liver, kidneys and spleen were not palpable. There were no deformities or edema of the extremities.

The diarrhea was controlled somewhat by the administration of an antidiarrhea mixture but his abdominal complaints persisted. On August 12, 1957, pitting edema of his legs was noted for the first time. No intravenous fluids and no rauwolfia medications [*20*] which might have resulted in fluid retention were given prior to the appearance of the edema. The total serum proteins were 4.0 gm. per cent; albumin, 2.0 gm. per cent; and globulin, 2.0 gm. per cent. The edema subsided by August 19, 1957, with no specific treatment except for a high protein, high caloric diet, and the patient was discharged. He was readmitted on August 31, 1957, with the chief complaint of swelling of both feet and legs. He also had abdominal cramps, nausea, vomiting and diarrhea, especially after eating. There was pitting edema of both legs and the sacral region. The total serum proteins were now 3.3 gm. per cent, with an albumin level of 2.9 gm. per cent and a globulin level of 0.4 gm. per cent. Two weeks later the total proteins were 3.5 gm. per cent, with an albumin level of 3.1 gm. per cent and a globulin level of 0.4 gm. per cent. The edema subsided and the patient was discharged on September 16, 1957. He later appeared in the medical clinic complaining of abdominal cramps aggravated by eating, and occasional loose bowel movements. There was some anorexia and nausea but no edema. The serum proteins were now normal for our laboratory: total proteins, 6.4 gm. per cent; albumin, 4.7 gm. per cent; and globulin, 1.7 gm. per cent. He responded to a regimen consisting of a bland diet, tincture of belladonna and Gelusil.® Only the bland diet was continued. He noted that

* From the Irwin U.S. Army Hospital, Fort Riley, Kansas.

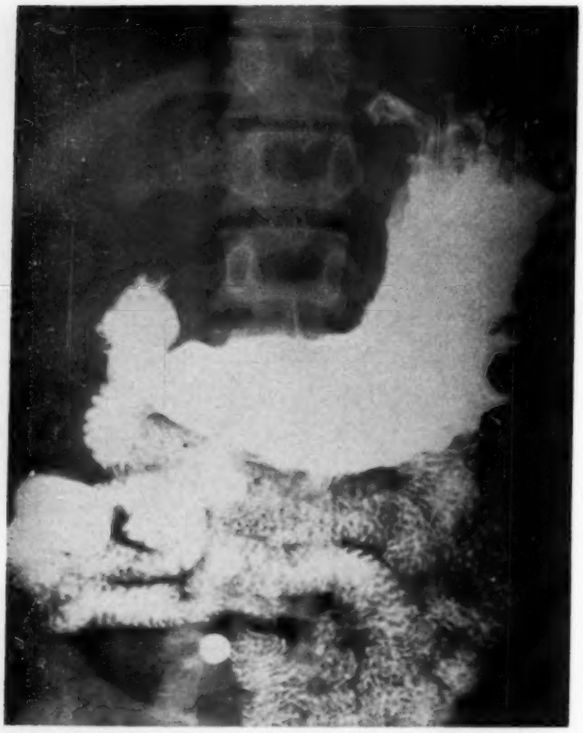

FIG. 1. Note the large rugal folds along the proximal greater curvature and similar but less prominent irregularities along the lesser curvature. These findings remained unchanged on all films.

when he failed to adhere to the diet his epigastric pain would recur.

On November 21, 1957, an examination of the upper gastrointestinal tract was carried out. At fluoroscopy large gastric rugal folds were noted and these were particularly pronounced along the greater curvature. (Fig. 1.) They were thought to be markedly in excess of the usually prominent folds along the greater curvature. Peristalsis was markedly diminished and the stomach lacked normal plasticity. On rotating the patient into the right anterior oblique projection a large filling defect was noted in the posterior wall of the stomach. (Fig. 2.) This had a sharp superior and

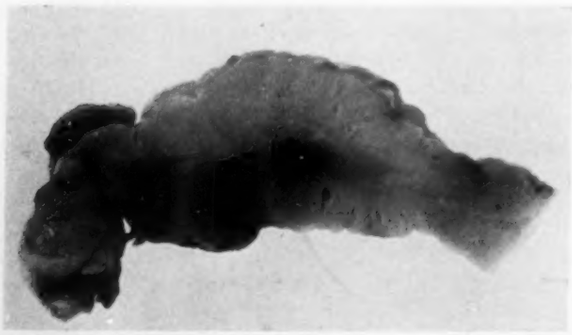

FIG. 3. The biopsy specimen. The mucosa is thickened and four intramucosal macroscopic cysts are evident.

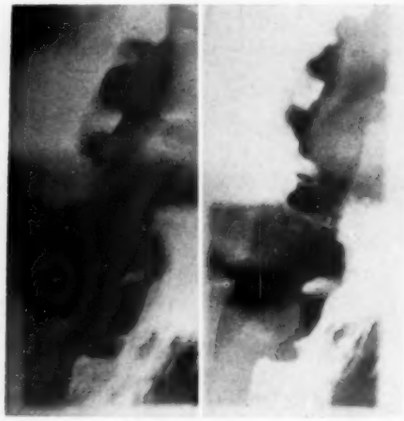

FIG. 2. Spot radiographs show a large filling defect on the posterior wall. It has sharp superior and inferior margins. The central barium collection suggests an ulcer, but this apparently represents barium trapped between rugal folds.

inferior border and revealed a central collection of barium thought to represent an ulcer crater. The roentgenological impression was lymphosarcoma of the stomach or giant gastric rugae. Radiological examination of the heart and lungs as well as an examination of the small and large bowel revealed no abnormalities.

Other laboratory data included a blood urea nitrogen of 11.4 mg. per cent and a blood creatinine of 1.25 mg. per cent. Repeated stool cultures and examinations for ova and parasites were negative. Serum agglutinins for typhoid O and H, paratyphoid A and B, brucella and shigella antigens were absent. Urine samples were of high specific gravity (maximum 1.029) and examination was negative for formed elements and albumin with the exception of 9 to 15 red blood cells per high power field in one specimen. A white blood count was 6,650 per cu.mm. with 66 per cent polymorphonuclear leukocytes, 30 per cent lympocytes, 1 per cent monocytes, and 3 per cent eosinophils. The hemoglobin was 15.1 gm. per cent. Liver function studies indicated a thymol turbidity of 2.1 units, a negative cephalin flocculation test, a total serum bilirubin of 0.52 mg. per cent, with 0.1 mg. direct and 0.42 mg. per cent indirect bilirubin, and an alkaline phosphatase of 0.72 units. Erythrocyte sedimentation rates were 1 and 2 mm. at the end of one hour.

No free hydrochloric acid was present after histamine stimulation. The patient experienced flushing and tachycardia after the injection of histamine, an indication that the stimulation was maximal.

Exploratory laporatomy was performed by Major James W. Cherry, M.C. At surgery the serosa of the stomach appeared normal. The wall was not thickened or indurated on palpation but was soft and boggy in consistency and had the so-called "bag of worms" feel. A 10.0 cm. gastrotomy was made in the anterior

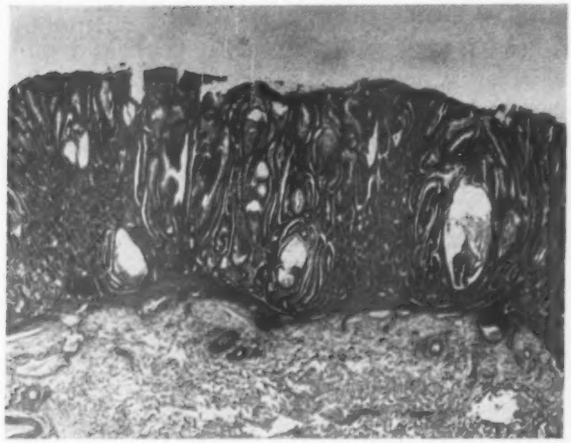

FIG. 4. Low power photomicrograph of the mucosa, muscularis mucosae and superficial submucosa. The gastric pits are elongated, tortuous and focally dilated. The entire thickness of mucosa in the center of the picture is composed of hyperplastic mucous cell elements. There is a very slight chronic inflammatory exudate. Original magnification × 32, enlarged about 2 diameters.

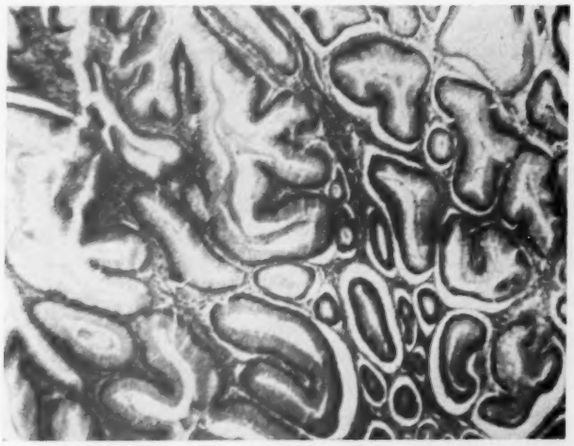

FIG. 5. High power photomicrograph showing tortuous branching crypts with focal dilatations. The lining cells are hypertrophied. Original magnification × 210, enlarged about 4 diameters.

wall of the pars media. Giant mucosal folds measuring up to 4.0 cm. in height covered the entire fundus and upper portion of the body. The mucosal folds revealed no ulcers or other gross lesions. The mucosa of the pyloric antrum and canal were normal. A biopsy specimen was taken from the margin of the gastrotomy.

On gross examination the specimen was an ellipse of stomach having over-all dimensions of 5.0, 3.0 and 1.0 cm. The serosa was smooth, pink and glistening. The mucosal aspect was flat, yellow-grey and slightly pebbled. Multiple sections revealed a normal muscularis and a loose, soft, grey-white submucosa. The mucosa was thickened, measuring between 3.0 and 4.0 mm. (Fig. 3.) Numerous macroscopic cysts measuring up to 0.5 mm. were noted in all layers of the mucosa. (Fig. 3.)

On microscopic examination the most striking feature was intramucosal mucus cell hyperplasia. (Fig. 4.) This was characterized by elongation, branching and tortuosity of the foveolae. (Fig. 5.) The cysts were focal dilatations of these pits, and contained mucinous secretion. The lining cells were columnar in type and very tall, with basally situated oval nuclei with stippled chromatin. Mitoses were frequent. The cytoplasm was eosinophilic and homogeneous. There was no atypism of the cells and no penetration of the muscularis mucosae. The gastric glands were reduced in number. (Fig. 4.) The inflammatory changes were minimal. Occasional nests of lymphocytes were found in the mucosa and superficial submucosa and a few scattered plasma cells, lymphocytes and eosinophils were noted throughout the mucosa. The submucosal vessels were dilated and congested (Fig. 4) and the surrounding connective tissues were slightly edematous. The muscle layers and serosa were normal.

COMMENTS

Giant hypertrophy of the gastric mucosa is histologically distinct and should not be confused with large but otherwise normal rugae or with chronic gastritis.

Large but otherwise normal rugae, simulating tumor of the stomach, have been repeatedly described [*12,18,23*]. Forssell has shown that the muscularis propria of the stomach determines the external form and width of the digestive tube but that the mucous membrane is freely movable against it, the muscularis mucosae being its special contractile organ. Varying with the stage of contraction of the muscularis propria, the mucous membrane may have an even surface or may show marked folding [*11*]. Spasm of the muscularis mucosae has been presumed to be the cause of large, otherwise normal rugae. Palpation of the antrum and pylorus of one patient at operation gave a recurrent sensation of induration with each spastic contraction of the prepyloric region [*23*]. Histologically the gastric mucosa was normal.

Patients with large but otherwise normal rugae may have gastrointestinal complaints. One elderly patient [*18*] had achlorhydria after histamine stimulation and some weight loss, but severe constitutional effects such as cachexia or hypoproteinemia have not been reported in these cases.

Giant hypertrophy of the gastric mucosa is a rare pathological condition. The mucous membrane of the stomach undergoes hypertrophy and hyperplasia in a zone that develops into a well

demarcated plaque composed of closely placed folds of mucosa [*2*]. The enlarged rugae appear to ride freely over the submucosa. The broadened, heavy, tortuous mucosal folds are crowded together to resemble convolutions of the brain. The mucous membrane at the edge of the plaque is normal. It may be thickened [*2*] or atrophic [*14*] at the base of the rugae and in the clefts between the rugae. The mucosa may vary from a pink [*1,8,14*] to a violaceous color [*1*] and may be edematous or hyperemic, and have ulcerations [*14,15*] or hemorrhagic spots [*9*].

The extent of involvement may vary from a localized hypertrophic ruga, which appears radiologically as a polyp [*2*], to a diffuse change involving the entire stomach. There is a predilection for the greater curvature [*1,2*]; the lesser curvature [*3*] is less frequently involved.

The histologic picture varies. The glands comprising the thickened mucosa may be greatly elongated and tortuous, with coiling to give the appearance of a corkscrew. Mucous cells, as well as chief and acid-secreting parietal cells, may be well differentiated and in normal proportions [*2*]. The gastric mucosa may be thickened as a result of elongation and hypertrophy of the upper mucous portions of the gastric glands and proliferation of the mucous epithelium lining the gastric surface and pits, with replacement of the acid-secreting and pepsinogenic cells [*13*]. In extreme cases there may be no chief or parietal cells [*13*]. The entire gland may be converted into a mucous crypt. In addition "metaplasia" of the epithelium with the appearance of goblet cells may occur. A grossly redundant and apparently hypertrophic mucosa may on microscopic examination resemble intestinal rather than normal gastric epithelium. There may be combinations of these variants of histology.

Retention cysts may be found. They are more commonly located in the basilar portions of the glandular layer but at times are present in the mid-mucosa, near the summit, or penetrate [*14*] the submucosa. These cysts are lined with a single layer of cuboidal or tall columnar mucus-producing cells [*1,2*].

Increased chromatism of nuclei and disorderly crowded nuclei may be present [*1*]. The interstitial tissue has a variable cellular infiltration, at times appearing normal and at times markedly inflamed. The muscularis mucosae is frequently thickened and often shows edema and mild inflammation. There may be benign glandular penetration of the muscularis mucosae [*1,2*]. The submucosa characteristically shows edema and an increase in vascularity. The muscularis propria at times is somewhat fibrotic and sometimes is somewhat thickened and edematous [*1,2*]. The serosa often exhibits unusually prominent vessels [*1*].

The cause of this condition is unknown [*2*]. Many hypotheses have been presented, including the possibility of a congenital anomaly or of the effects of chemical, thermal or mechanical irritants or infectious agents [*2*].

The roentgenological manifestations vary with the extent of the lesion. When the disease is localized the radiographic studies may reveal only a single filling defect similar to a polyp [*2*] or a nodular tumour. When the lesion is more extensive it may be confused radiologically with lymphoma, polypoid carcinoma, diffuse submucosal carinoma and multiple polyposis. In rare instances the stomach has been found to lack normal plasticity to the radiologist's palpating hand [*1*].

The differentiation of large but otherwise normal rugae from true mucosal hypertrophy is difficult without adequate tissue examination. The reduction of the gastric folds to normal size with the subsidence of peristalsis and/or air insufflation suggests large but otherwise normal rugae, as with Ricketts' patients in whom biopsy was not obtained [*12*]. These features may be lacking [*23*], however, and histologic study is mandatory. Belladonna and atropine usually have no effect upon these rugae [*18,23*]. The reason for this is not clear.

The literature is confusing, partly because large rugae with a histologically normal mucosa are called hypertrophic, and partly because roentgenologists are in the habit of labeling large rugae hypertrophic merely because of their size [*21*]. Furthermore, inflamation of the mucosa may be absent [*2*] or severe [*1*]. Gastritis is occasionally diagnosed.

Clinical Picture. The clinical picture is variable and not diagnostic. No gastric complaints may be present: two patients sought medical care because of sore gums and a ureteral stone, respectively [*2*].

Ulcer-like upper abdominal pain, often completely relieved by the ingestion of food and alkalies is frequently present. However, the pain may be diffuse and crampy or described as a feeling of pressure or burning and may be aggravated by food. Belching, nausea and vomiting occur less frequently. Acute pyloric obstruction

because of the redundant mucosa has been demonstrated at operation [*22*] and is one cause of vomiting. Constipation and diarrhea are less often present. Hematemesis and/or melena may occur and occasionally they have necessitated gastrectomy [*16,19*]. Physical examination of the abdomen is usually within normal limits, except for occasional cases in which epigastric tenderness is present [*16*]. Associated duodenal ulcer may be present [*1,2*].

Peripheral edema may cause the patient to seek medical attention. The appearance of hypoproteinemia and edema is not always related to the presence or severity of gastrointestinal symptoms. Easy fatiguability and edema of the legs were the presenting complaints in Case II of Spellberg [*9*]. In Citrin's case the marked hypoproteinemia and edema were evident one year prior to the development of gastrointestinal symptoms [*8*]. However, Balfour's patient had intractable abdominal complaints, with hypoproteinemia and edema, and responded only to total gastrectomy [*10*]. The hypertrophied mucosa had a normal complement of mucous, chief and parietal cells with focal areas of mucous cell hyperplasia and cyst formation [*10*].

Weight loss is frequently present and may be severe, but may be absent. Obesity because of a concurrent hypothalamic lesion has been described [*2*].

Constitutional symptoms of varying severity may be present for which there is no explanation. Schindler et al. described a patient with extraordinary cachexia, a progressive downhill course, and early death. The patient had diarrhea, occult blood in the stools and anemia. The suspicion of malignancy was not verified at autopsy; instead, a thickened gastric mucosa consisting almost entirely of a proliferation of the upper mucous portions of the gastric glands and surface epithelium, which was devoid of gastric glands, was found [*13*]. A similar histologic picture was present in Case IV of Maimon et al. but the patient did not have progressive cachexia. Death was due to embolization (this case was erroneously cited in a recent report [*8*] as one of severe cachexia and death due to giant hypertrophy of the gastric mucosa). Evidence of constitutional effects of lesser severity were evident in Case I of Spellberg in which the patient sought medical advice because of weight loss and fatiguability, in the absence of anemia or severe hypoproteinemia [*9*].

Multiple endocrine adenomas were found in three patients (pituitary adenomas, islet cell adenomas, parathyroid tumors [*2*]). The relationship to the gastric lesion is not known and may be entirely coincidental. Two subjects had associated clubbing of the fingers [*5,6*].

The duration of symptoms has varied from two months to twenty-two years with an average duration of two years [*2*].

There have been no reported cases, as such, in adolescents or children. However, one three year old patient reported as having had "idiopathic hypoproteinemia" had large gastric rugal folds and is probably the youngest patient with this disorder described in the literature [*24*].

Achlorhydria after histamine stimulation may be present, especially in patients who have suffered replacement of the acid-secreting cells by the proliferative mucosa [*1*]. In one series of patients with adequate parietal cells on histologic examination an abundance of free hydrochloric acid was present. In some of these patients achlorhydria was present but histamine was not used [*2*]. Anemia is only occasionally found.

Giant hypertrophy of the gastric mucosa is not generally recognized as a cause of edema or of "idiopathic hypoproteinemia." No instance of unexplained edema should be considered completely evaluated without a careful study of the gastric mucosa. Large rugae, unless specifically looked for, may be overlooked on a routine roentgenological examination of the stomach.

The hypoproteinemia in giant hypertrophy of the gastric mucosa may occur with great rapidity, even with an adequate protein and caloric intake. In Case II of Maimon et al. there was a fall in serum proteins from 5.1 gm. per cent to 3.8 gm. per cent in sixteen days [*1*]. Citrin et al., using I^{131}-labeled albumin, have elucidated the probable pathogenesis of the hypoproteinemia. Their subject had a markedly accelerated turnover of albumin compared to normal control subjects. With a total exchangeable albumin pool less than one-half that of normal control subjects, the turnover was so rapid that the absolute degradation rate in grams per day was approximately equal to that of the control subjects. They found that the gastric juice contained large amounts of albumin, accounting for almost one-half of the patient's daily albumin loss [*8*]. Thickening of the gastric mucosa in their patient was a result of overgrowth of the mucous type of epithelium.

The existence of this lesion in a three year old

child suggests the possibility of a congenital lesion but an acquired cause cannot be excluded.

The patient described in this paper was a diagnostic problem, initially because of unexplained hypoproteinemia and extensive edema, and subsequently because of the suspicion of gastric malignancy. When gastric rugal folds of the prominence noted in the present case are encountered the possibility of malignancy cannot be excluded by roentgenographic studies. Loss of plasticity of the stomach to the radiologist's palpating hand and the presence of the filling defect were so suggestive of a malignant neoplasm in the present instance that an exploratory laporatomy was undertaken, only to find giant rugal folds which histologically were diagnostic of giant hypertrophy of the gastric mucosa.

In the individual case adequate tissue examination is the only certain way to diagnose this lesion and to exclude malignancy. A surgeon's palpating hand has, in individual instances, been unreliable in excluding malignancy. The patient with constant giant rugae may be gastroscoped and a biopsy taken if a competent gastroscopist is available.

If, after all factors have been evaluated, malignancy cannot be excluded, exploratory laporatomy with biopsy is indicated. A localized lesion should be resected. The majority of cases may be managed conservatively. Only when the symptoms are intractable or hemorrhage is uncontrollable (and the lesion diffuse) is a subtotal gastrectomy indicated. Symptoms are relieved in most instances even though the rugae are incompletely removed [*9,16*]. If the lesion is localized to the proximal portion of the stomach subtotal gastrectomy is of no value. Removal of the proximal portion of the stomach or total gastrectomy [*17*] should be reserved for subjects in whom all other measures have failed.

SUMMARY

A twenty-five year old Negro man, a diagnostic problem initially because of unexplained hypoproteinemia and edema, and subsequently because of the suspicion of gastric malignancy, was found to have giant hypertrophy of the gastric mucosa.

The clinical picture, radiological manifestations, differential diagnosis, pathology and management of this condition are discussed. Differentiation from large rugae with normal mucosa is stressed. Gastric mucosal study may disclose the diagnosis in instances of unexplained edema and/or hypoproteinemia.

Acknowledgment: We express our appreciation to Major James W. Cherry, M.C. and Capt. Myron D. George, M.C. for their assistance in managing this patient and to Capt. John E. Rogers, Jr., M.S.C. for technical assistance.

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Hemolytic Anemia Induced by Sulfoxone Therapy, with Investigations into the Mechanisms of Its Production*

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THE large number of chemical agents to which persons are being exposed at the present time has brought to light certain hematopoietic sensitivities which may lead to serious hematological disease in the presence of an offending substance. That there are agents of the benzol family which lead to profound myeloid disease has been known for many years [*1*]. Many drugs have also been recognized as hemolytic agents [*2–4*], and Alving et al. have recently described some of the biochemical abnormalities in the erythrocytes of a group of the population susceptible to the hemolytic effects of these drugs [*5–8*].

This report describes the induction of hemolytic anemia by one of these agents in a patient in whom there was no evidence of such a biochemical defect.

METHODS

The methods for routine blood studies as well as for assays of glutathione sensitivity, glucose-6-phosphate dehydrogenase and methemoglobin reductase have already been described in detail [*9*]; 6-phosphogluconate dehydrogenase activity was calculated by observing the rate of reduction of triphosphopyridine nucleotide as measured by changes in optical density at 340 lambda using silica cells in a Beckman DU spectrophotometer with a hydrogen lamp light source. The incubation mixture consisted of 15 μmoles of 6-phosphogluconate, 4 μmoles of triphosphopyridine nucleotide (TPN), dialysed hemolysate equivalent to 0.1 gm. per cent of hemoglobin in the final reaction mixture, and Tris buffer, 0.036 molar in a volume of 3 ml. Glutathione reductase was measured by observing the amount of reduced glutathione produced in fifteen minutes in an incubation mixture of 3.5 ml. containing 2.5 μmoles of oxidized glutathione, 1.6 μmoles of reduced triphosphopyridine nucleotide, dialyzed hemolysate containing 2 mg. of total heme pigment, 25 μmoles of EDTA and Tris buffer to a final concentration of 0.2 molar. Glutathione was measured as described by Grunert and Phillips [*10*] after stopping the reaction by the addition of 4.5 ml. of 3.33 per cent metaphosphoric acid. Glutathione sensitivity was measured according to the technic of Beutler [*5*]. Iron incorporation studies were carried out with Fe^{59} containing a specific activity of more than 1 μc. per μg. The plasma was tagged and samples were drawn and counted in a Tracerlab well with a 3-inch lead shield as previously described [*11*]. Surface monitoring was performed with a Tracerlab scintillation probe, with a 1-inch window and a 2-inch lead shield projecting two inches beyond the window. This was held flush against the marked site with the patient in the same position throughout the serial readings. The background of this probe was 170 counts per minute.

CASE REPORT

The patient was a fifty year old man in whom methemoglobinemia and hemolytic anemia developed while receiving sulfoxone therapy for Hansen's disease. He had clinical evidence of leprous involvement of the ulnar nerve, confirmed by biopsy of a granuloma. Before treatment the hematocrit was 48 to 50 per cent and red cells appeared normal on smear. He was first given 150 mg. of sulfoxone daily, with a gradual increase of the dose over the next nine weeks to 1.2 gm. daily. Approximately four months after treatment began the patient took 2.4 gm. daily in error and thereupon nausea, weakness and malaise developed. When seen three weeks later it was apparent that the patient's skin had a cyanotic hue; laboratory studies revealed a mild anemia.

At this time the blood findings were as follows:

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hemoglobin, 12.0 gm. per cent; hematocrit, 42 per cent; reticulocytes, 7.3 per cent; white cells and differential count, normal; and marrow, normal with M:E ratio of 1.6:1. Seventeen per cent of the total heme pigment was methemoglobin. The patient was also found to be slightly deficient in iron as evidenced by erythrocyte indexes indicating hypochromia, and a serum iron drawn after cessation of hemolysis was 50 μg. per cent. Administration of sulfoxone was omitted for six weeks in order to evaluate the patient's normal values. Methemoglobinemia disappeared and the hematocrit rose to a level of 48 to 51 per cent.

EXPERIMENTAL STUDIES

In order to evaluate the effect of the drug on aging cells, the patient was given normal plasma tagged with radioactive iron, which was incorporated completely into his red cells, thereby producing a labelled population of a single age group. (Fig. 1.) After the red cell radioactivity reached a plateau, he received 1 gm. of ferrous sulfate daily in divided doses for the duration of the experiment to inhibit reutilization of the radioactive tag. There was a slight reticulocyte response to this therapy with some elevation in hemoglobin as well. Surface monitoring of sacrum, precordium, liver and spleen demonstrated rapid entrance and exit of the tag to and from the marrow, with return to the circulation followed by a steady state over the organ sites. At this time the patient was given a daily dose of 1.2 gm. of sulfoxone. Methemoglobinemia reappeared, together with a fall in hematocrit and an increase in reticulocytes; however, this was not associated with hemolysis of the tagged young cell population. After one week, treatment was withdrawn for three days because of nausea and malaise, then reinstituted at the thirty-sixth day. After the forty-ninth day from administration of the radioactive tag, there was disappearance of the tagged cells from the circulation. At this time, surface scanning revealed increase in radioactivity over the spleen, probably reflecting the localization of erythrocyte destruction. The patient compensated for hemolysis with a reticulocytosis and maintained approximately 5 per cent circulating methemoglobin.

Investigation of methemoglobin reductase activity in the erythrocytes of this patient was carried out by observing the rate of reduction of methemoglobin on incubation with glucose or lactate in concentrations of physiologically equivalent molarity. These studies of methemoglobin reduction through the DPNH pathway derived *in vitro* from lactic dehydrogenase demonstrated normal rates. The acceleration of the reduction of methemoglobin upon addition of methylene blue to glucose, probably related to the production of TPNH in the hexose monophosphate shunt, was also within normal limits. Thus there was no evidence of abnormal methemoglobin reduction by either of the known methemoglobin reductase systems. In Table 1 the results of these experiments are given.

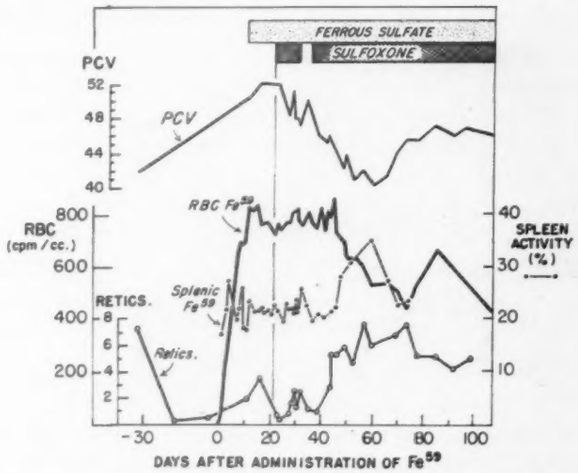

FIG. 1. The effect of sulfoxone on a tagged population of red cells of similar age is demonstrated. PCV refers to packed cell volume. Red cell radioactivity is expressed as counts per minute per milliliter of cells and is plotted according to days after administration of Fe^{59} with correction for physical decay. Splenic radioactivity is expressed as percentage of total activity noted over standard monitoring sites. It is apparent that hemolysis of the general population, as evidenced by a decline in packed cell volume associated with reticulocytosis, occurred before the tagged cells were hemolyzed.

Since this patient showed evidence of hemolysis as well as methemoglobinemia, investigations were carried out to determine whether or not the patient had a biochemical defect of his red cells similar to that found in patients sensitive to primaquine. Glutathione stability was tested in the usual manner by observing the reduced glutathione content of the red cells after two and four hours' incubation with acetylphenylhydrazine. The results were normal, and the curve is plotted in Figure 2.

Further studies were carried out to investigate the enzyme systems involved in the maintenance of glutathione in the reduced state. Assays were made of glucose-6-phosphate dehydrogenase and 6-phosphogluconic dehydrogenase, the two sources of TPNH in the glycolytic scheme

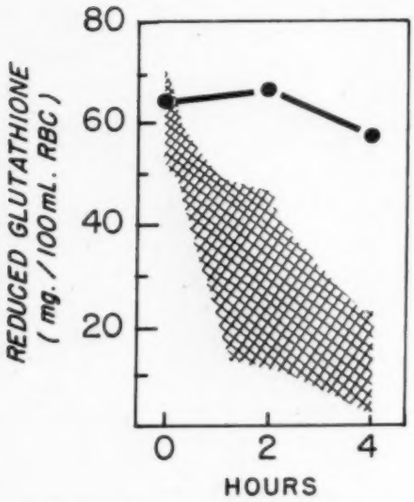

FIG. 2. The reduced glutathione of the patient's red cells is plotted against hours of incubation *in vitro* with acetylphenylhydrazine. The shaded area represents curves found in the blood of patients who have glutathione "sensitivity." The patient's curve is normal.

in the erythrocyte, and also of glutathione reductase, which depends on TPNH generated by these two previous enzymes. The two former enzymes were found to be within normal limits, while the latter was diminished. The results are presented in Table II. It would thus appear that this patient does not have the enzymatic deficiency described by Alving et al., characterized by a decrease of glucose-6-phosphate dehydrogenase associated with an increase of glutathione reductase [*7,8*].

COMMENTS

Drugs which lead to hemolytic anemia coincident with interference with glutathione reduction are of the "oxidant" group and probably inhibit one or more enzyme systems involved in hydrogen transfer. This group includes such commonly used medications as phenacetin, nitrofurantoin and sulfanilamide; sulfoxone has also been shown to have *in vitro* effects similar to this group of medications in susceptible patients [*4*].

The incidence of anemia in patients receiving sulfoxone has been found to be extremely high; and it is a common clinical observation that such patients have an early drop in hemoglobin followed by a partial recovery with equilibration at that level [*12*]. The effect of sulfoxone appears to be related to dosage and is cumulative. This may explain the lack of hemolysis noted in normal subjects in short-term experiments in

TABLE I
IN VITRO REDUCTION OF METHEMOGLOBIN BY PATIENT'S ERYTHROCYTES

Glucose	Glucose and Methylene Blue	Lactate	Lactate and Methylene Blue	PO_4 Saline	PO_4 Saline and Methylene Blue
Methemoglobin Reduction in 180 Minutes					
(0.6–25) 3.8	(6.1–8.8) 7.4	(0.0–2.5) 1.4	(0.8–3.8) 1.7	(0.0–0.8) 0.3	(0.0–1.3) 0.4
Methemoglobin Reduction in 300 Minutes					
(1.5–6.8) 5.2	(5.6–10.5) 8.3	(1.3–4.5) 2.1	(1.9–6.8) 2.7	(0.0–2.3) 0.2	(0.0–2.9) 0.8

NOTE: The amount of methemoglobin reduced by the patient's red cells in the presence of the designated substrate is expressed per 10 gm. of hemoglobin present. Observations after 180 minutes incubation and after 300 minutes incubation are compared to the normal range which is presented in parentheses under each substrate. These results show no evidence of deficiency of the methemoglobin reduction systems.

which 0.3 gm. of sulfoxone was given daily for eight days [*4*], and up to 0.6 gm. for twenty-eight days [*13*]. Rarely, severe hemolysis occurs, and one might speculate that it is in this latter situation that a congenital enzymatic deficiency of glucose-6-phosphate dehydrogenase allows an exaggeration of the toxic effect of the drug [*14*].

In the case reported here, there was a slight deficiency of glutathione reductase activity. This was not of a degree to cause an abnormal glutathione sensitivity test, and that it could have *in vivo* effects in the presence of large doses of the drug can only be suggested. In view of the

TABLE II
ENZYME ACTIVITY OF PATIENT'S ERYTHROCYTES

Enzyme Activity	Values	Normal Range
Glucose-6-phosphate dehydrogenase	.050	.040–.096 O.D.*
6-Phosphogluconic dehydrogenase	.062	.030–.070 O.D.
Glutathione reductase	20 mg.	28–39 mg.

NOTE: The results of assays of enzyme activities of hemolysates of the patient's blood are compared to the normal range. Glucose-6-phosphate dehydrogenase activity and 6-phosphogluconic dehydrogenase activity are expressed as average five-minute increments in optical density at 340 lambda. Glutathione reductase is expressed as milligrams of glutathione reduced. The results demonstrate normal values for the former two enzymes and diminished activity for the glutathione reductase.

* O.D. = optical density.

fact that anemia is said to be such a frequent occurrence and is related to dose [*15*], it is probable that hemolytic anemia occurs with this particular drug even in the presence of normal enzymatic activity.

The importance of glutathione to viability of the red cell has been suggested by several experimental and clinical observations. Sheets and co-workers have shown that sulfhydryl inhibitors increase osmotic fragility *in vitro*, a reaction that can be prevened by the presence of glutathione in sufficient amount [*16*]. Benesch and Benesch have demonstrated that oxidation of glutathione *in vitro* will actually lead to autohemolysis [*17*]. Studies in patients sensitive to primaquine [*6,7*], in patients with fava bean hemolysis [*18*], and in a series of cases with hemolytic anemia due to naphthalene [*9,19*] give clinical support to the theory that glutathione reduction is necessary for erythrocyte integrity. Although its actual function in the red cell is not clearly understood, glutathione is known to have a rapid turnover in adult erythrocytes, and may take part in several types of metabolic reactions upon which erythrocyte metabolism depends.

The observance of hemolysis of the tagged cell population only after approximately fifty days had been spent in the circulation suggests that hemolysis is a phenomenon reflecting the increased susceptibility of the aging cells to this agent. Such an interpretation is consistent with evidence that both glucose-6-phosphate dehydrogenase and glutathione reductase decrease with the age of the cell [*20,21*]. This susceptibility allows equilibration of hemoglobin levels by marrow compensation to a shorter red cell age span, and explains the clinical "recovery" in these patients when sulfoxone is continued. This course is the same as noted in the primaquine-sensitive group given drugs [*4*].

The prominence of methemoglobinemia in the case presented has not been noted in most drug-induced hemolytic anemias and one cannot say whether this is a parallel abnormality or related in some way to the hemolytic mechanism. Sulfoxone is known to lead to methemoglobinemia with great frequency, and it has been observed clinically that the methemoglobinemia decreases as the patient is kept on the drug for a longer time [*22*]. Such a course is consistent with destruction of older cells containing less adequate enzyme systems for methemoglobin reduction, with a resulting compensated hemolytic anemia and a younger cell population.

Ponder has suggested that the presence of methemoglobin in sufficient amounts may, in itself, lead to stromal changes and hemolysis [*23*]. However, there is no evidence that erythrocytes of congenital methemoglobinemia hemolyze more rapidly than normal, and since the patient had less methemoglobin than the amount seen in the congenital disease, it is unlikely that the pigment was of itself responsible for the hemolytic anemia.

SUMMARY

A case report is presented of a patient in whom clinical evidence of hemolytic anemia with methemoglobinemia developed upon increasing the dosage of sulfoxone which he was receiving for Hansen's disease. Administration of the drug was withdrawn for six weeks, after which studies were made with the use of radioactive iron to demonstrate the hemolytic effect of sulfoxone. By means of tagging cells of a single age group, it was shown that the susceptibility to hemolysis was related to cell age, and it was only after the red cells had been in the circulation for approximately fifty days that they were subject to the hemolytic effects of this drug.

No significant biochemical defect could be demonstrated in the erythrocytes of this patient. However, the decline in activity of glucose-6-phosphate dehydrogenase and glutathione reductase in the aging red cell and the reported sensitivity to this drug of patients with red cell deficiency of glucose-6-phosphate dehydrogenase suggest that the hemolytic mechanism is related to this enzyme system. The case demonstrates that drug-induced hemolytic anemia of this type may occur without demonstrable enzymatic defect of the erythrocytes.

The observation that patients appear to compensate for the hemolytic anemia and methemoglobinemia if maintained on this drug may be explained by the fact that the older cells containing less adequate enzyme systems are removed from the circulation and an equilibrium is maintained by increased production of red cells with a shorter age span.

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Esophageal Varices in Hodgkin's Disease Involving the Liver*

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ALTHOUGH the recorded death rate due to Hodgkin's disease in the United States per 100,000 of the population was 1.7 in 1951 [*1*] (and it is claimed that the liver is involved with Hodgkin's disease in about 50 per cent of the cases [*2–4*]), to our knowledge no case of esophageal varices due to intrahepatic portal hypertension caused by Hodgkin's disease of the liver has been reported. In a study of the liver in Hodgkin's disease which is now in progress [*27*], we noted two patients with Hodgkin's disease and accompanying liver involvement who had esophageal varices. In one of the patients the only demonstrable cause for the esophageal varices was extensive intrahepatic Hodgkin's disease. In the other, in addition to liver involvement with Hodgkin's disease, fibrinous constrictive pericarditis was present. Both patients had gastrointestinal hemorrhage. We believe that a presentation of these two cases is pertinent.

CASE REPORTS

CASE 1. R. G., a thirty-seven year old white male pilot, first consulted one of us (L. F. C.) on October 13, 1949. His present illness had started on April 10, 1948, when he noted a mass in his right axilla and a decrease in vigor. The diagnosis of Hodgkin's disease had been established at another institution† by lymph node biopsy on May 24, 1948; he had been given x-ray therapy to the right axillary and supraclavicular areas.

His complaints when first seen at Memorial Center were weakness and pain in the low mid-back. The physical examination revealed no abnormalities, but roentgenograms of the pelvis showed a punched-out bony defect in the right ilium. Peripheral blood examination was within normal limits. The bone lesion was treated with radiation therapy.

† Confirmed also at Memorial Center.

Following this therapy the patient felt well until July 1949 when he complained of pain, fever and weight loss. On September 4, 1949, a tender, left iliac mass was found, and another mass was felt in the left para-aortic region. X-ray films of the pelvis revealed no interval change, and the peripheral blood examination was still within normal limits. Radiotherapy to the abdomen was started through four ports. Following the thirteenth treatment on October 3, 1949, severe abdominal pain, vomiting, and right lower abdominal spasm and tenderness developed.

Exploratory celiotomy with the patient under spinal anesthesia was performed by Dr. Lemuel Bowden on October 4, 1949. The surgical findings were an acutely inflamed appendix (histologically, acute suppurative appendicitis) and cloudy peritoneal fluid. The appendix was removed, but no further exploration for intra-abdominal Hodgkin's disease was attempted because of the peritonitis. The postoperative course was uneventful.

No further x-ray therapy was given to the abdomen, and the patient remained quite well for four weeks when he noted a nodal enlargement in the right side of the groin. Physical examination revealed a node, 4.5 cm., in the right side of the groin. This node was treated with radiotherapy, with good results. On November 2, 1949, the patient had high fever and chills accompanied by abdominal pain. The spleen became palpable for the first time; and when his condition did not improve, he was admitted to Memorial Center on November 22, 1949.

Physical examination on admission revealed a temperature of 102°F., pulse 96 per minute with regular sinus rhythm, and a blood pressure of 100/70 mm. Hg. A round node, 1 by 1 cm., was felt in the left side of the groin and another of the same size was located in the right side of the groin. In addition, a node, 3 by 2 cm., was felt high in the right axilla. The liver was palpable 8 cm. below the costal margin in the mid-clavicular line, had a sharp edge, and was slightly tender. The spleen was palpable 6 cm. below

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FIG. 1. Case 1. Spleen (top) and liver showing discrete nodules of Hodgkin's sarcoma.

the costal margin in the mid-clavicular line. The patient was clinically somewhat jaundiced.

Laboratory findings upon admission to the hospital were as follows: the peripheral blood showed a hemoglobin of 10.5 gm. per cent; white blood cell count, 1,000 per cu. mm.; and platelet count, 82,000 per cu. mm. A differential smear revealed polymorphonuclear cells, 60 per cent; non-filamented polymorphonuclear cells, 10 per cent; lymphocytes, 23 per cent; metamyelocytes, 4 per cent; and myelocytes, 1 per cent. The hematocrit was 35 per cent; reticulocytes, 2.4 per cent. Blood chemistry determinations revealed a total serum bilirubin of 2.4 mg. per cent; the cephalin flocculation test 1-plus in twenty-four hours; total serum protein, 5.3 gm. per cent; prothrombin time, normal; and blood urea nitrogen, 16.4 mg. per cent. The results of the Mazzini test were negative. No abnormalities were noted in the urine. A stool specimen gave a slightly positive reaction for occult blood.

A bone marrow aspiration showed the following: reticulum cells, 0.6 per cent; myelocytes, 33.5 per cent; metamyelocytes, 24.8 per cent; non-filamented forms, 26.6 per cent; filamented forms, 11 per cent; eosinophils, 0.5 per cent; lymphocytes, 3.6 per cent; plasma cells, 0.2 per cent; normoblasts, 24.6 per cent; and erythroblasts, 0.1 per cent.

X-ray films of the patient revealed elevation of the left diaphragm and the presence of bone destruction of the right ilium as previously described.

The patient was treated with x-ray radiation to the spleen. After completion of this therapy an elevation in hemoglobin level (to 13.0 gm. per cent) was noted. The white blood cell count and platelet count remained essentially unchanged. Following radiotherapy to the spleen a further increase in the size of the liver (up to 10 cm. below the costal margin in the mid-clavicular line) was noted.

Three days after completion of radiotherapy to the spleen the patient became confused, then semicomatose, and went into shock. Physical examination at this time revealed a marked increase in icterus, labored breathing, and peripheral cyanosis. The pulse and blood pressure were not obtainable. A peculiar odor (fetor hepaticus) was noted. The pupils were dilated but reacted slightly to light, and tendon reflexes could not be obtained. The patient succumbed on the day following the acute episode despite supportive treatment with intravenous fluids, neosynephrine and adrenocortical extracts. No hematemesis or melena preceded the acute episode. Laboratory studies were not obtained during the last hours of the patient's life.

The total duration of disease was twenty months from the first symptom, two months after radiation to the abdomen and appendectomy, and three days after completion of radiotherapy to the spleen.

The postmortem findings revealed the following: Macroscopically the liver was firm and weighed 2,750 gm. (Fig. 1.) The capsule was intact, smooth and glistening. The surface and cut section were mottled red and yellowish green. In the left lobe of the liver there were three grayish white areas, each measuring 3 cm. in size. The central portions of the lobules were red and the peripheral portions yellow-brown. The portal and hepatic veins were patent. Microscopic examination of the liver (Fig. 2) showed diffuse periportal Hodgkin's disease, marked atrophy of the liver cells, and moderate fatty infiltration. In addition, passive hyperemia and hemosiderosis were present.

The gallbladder was 7 cm. long and 1.5 cm. in diameter. Bile ducts were patent. Microscopically there was diffuse infiltration of Hodgkin's disease in the gallbladder, located mainly in the vessels.

The spleen weighed 1,430 gm. and was 17 by 15 by 9 cm. in size. Its surface was studded with multiple grayish whitish yellowish areas measuring up to 3 cm. in diameter. Numerous white and yellow areas were seen throughout the organ. (Fig. 1.) Microscopically Hodgkin's disease was seen in many areas, in the walls of the vessels, and in the lumina of large vessels. Hemorrhage, fibrosis, necrosis and a zone of infarction were also seen.

The mucosa of the upper two-thirds of the esophagus was pink, smooth and glistening. In the lower one-third it was hemorrhagic, with dilated, engorged veins. Microscopically (Fig. 3), ulceration and necrosis of the esophageal mucosal blood vessels were found.

There was also Hodgkin's sarcoma involving the lymph nodes, skin and bone marrow. A bluish green fluid was present in the stomach, and 2,000 cc. of a slightly opaque, yellowish brown ascitic fluid was found in the abdominal cavity. The pleural cavities each contained 200 cc. of yellow fluid, and pulmonary

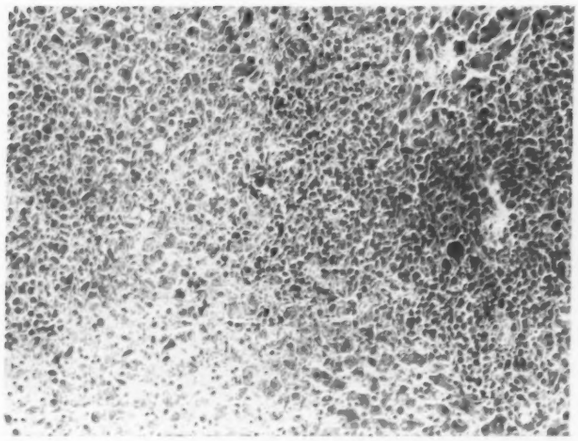

FIG. 2. Case I. Replacement of hepatic parenchyma with Hodgkin's sarcoma cells.

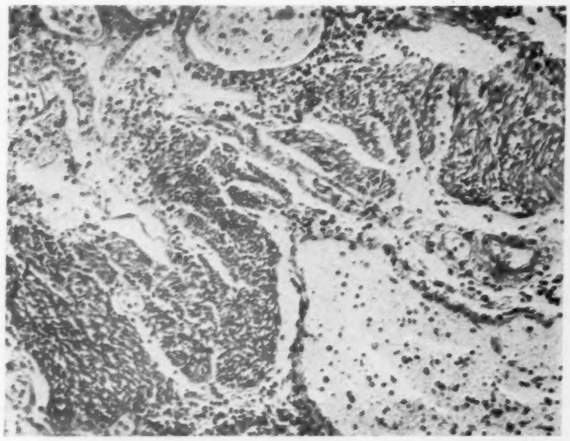

FIG. 3. Case I. Wall of esophagus showing large venous spaces in mucosa (top) and between muscle (lower right). The overlying surface epithelium was eroded. Inflammatory cells are present in the wall.

edema was present. Other pathological findings were bilateral compensatory emphysema, adrenal cortical adenoma, infarction of the spleen, multiple junctional nevi and testicular atrophy.

CASE II. M. B., a seventeen year old white boy, was first admitted to Memorial Center on January 27, 1955, with evidence of an early superior vena caval obstruction syndrome associated with cough of several weeks' duration. Evaluation during his first admission to Memorial Center revealed cervical and mediastinal lymphadenopathy with elevated venous pressure in the upper extremities. The diagnosis of Hodgkin's disease was established on biopsy of a cervical node, and he was treated with radiation therapy to the neck and mediastinum preceded by a full course of nitrogen mustard. He was discharged much improved and was followed as an outpatient.

It was not until one year after onset of the disease that his liver became palpable. At the same time he required further radiation therapy for symptoms and signs suggestive of recurrent superior vena caval obstruction syndrome. Because of pain in the back, a mass in the lower left side of the chest, and an enlarging liver, the patient was again admitted to the hospital on April 5, 1956. At this time the patient demonstrated a paradoxical pulse. A mediastinal mass was again demonstrated, and he was treated with nitrogen mustard followed by radiation therapy. The patient was discharged from the hospital before radiotherapy was completed, and he continued receiving his treatments on an outpatient basis since he was feeling much improved.

Twenty-two days later he was admitted for the third time because of fever, anorexia, dyspnea and non-productive cough. Once again signs of a full-blown superior vena caval compression syndrome were present. A para-aortic mass was palpable as well as generalized nodal enlargement. X-ray films of the patient's chest again revealed massive mediastinal nodes and an abnormal cardiac silhouette suggestive of pericardial disease.

The peripheral blood examination was within normal limits, urinalysis revealed normal findings, and the urinary output was adequate. Blood chemistry determinations revealed a blood urea nitrogen of 13.4 mg. per cent; total serum bilirubin, 0.52 mg. per cent; cephalin flocculation test, 2-plus in twenty-four hours; thymol turbidity test, 0.16 units (normal range in our laboratory is up to 1.75 units); serum alkaline phosphatase, 3.1 Bodansky units; serum total protein, 7.7 gm. per cent; albumin, 4.6 gm. per cent; uric acid, 4.9 mg. per cent; calcium, 9.5 mg. per cent; and phosphorus, 3.3 mg. per cent. Results of both indirect and direct Coombs' tests were negative.

The patient responded to bedrest, nitrogen mustard and radiotherapy, and was discharged and followed up for the next five months on an outpatient basis. Initially he did quite well, but progressive weight loss, anorexia and fever later appeared. A partial collapse of the right lung also occurred. Triethylene melamine (TEM) was given orally for a period of two weeks, but no improvement was noted. Thereafter prednisone, 30 mg. daily, was started. One week later there was epigastric pain in the left upper quadrant of the abdomen. One day prior to his last hospital admission he had a rather sudden onset of shortness of breath and passed a black, tarry stool. This episode prompted his final hospital admission. The blood count had been within normal limits prior to this episode.

Physical examination revealed a temperature of 102°F.; pulse, 146 per minute and regular; blood pressure, 90/70 mm. Hg in both arms. The patient was pale and agitated as well as markedly dyspneic. The neck veins were slightly distended and did not pulsate, and the trachea was deviated to the right. Nodes were felt in the inframandibular, posterior

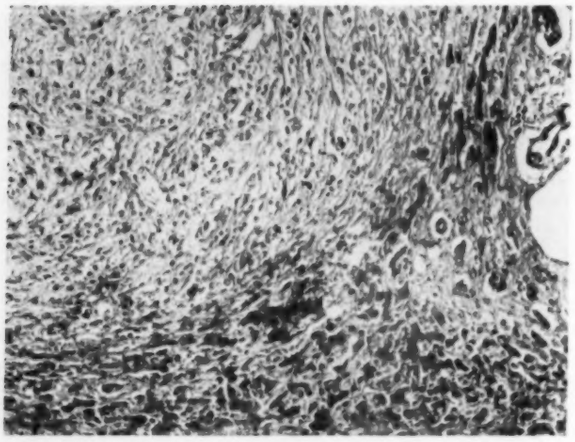

FIG. 4. Case II. Focus of Hodgkin's granuloma (markedly fibrotic) compressing hepatic tissue.

cervical, right axillary and inguinal areas. Widening of the superior mediastinum was found on percussion, and dullness to percussion was present at the right base. Wheezing over the entire right chest was heard. A systolic murmur, loudest at the base, was heard and transmitted to the apex. Liver and spleen were not palpable. Neurological examination was within normal limits.

Peripheral blood examination revealed the following: hemoglobin, 8.9 gm. per cent; hematocrit, 25 per cent; platelets, 168,200 per cu. mm.; leukocyte count, 9,700 per cu. mm.; polymorphonuclear leukocytes, 83 per cent; band forms, 4 per cent; eosinophils, 4 per cent; monocytes, 8 per cent; and lymphocytes, 1 per cent. Bleeding time was five minutes and clotting time was three minutes and fifty seconds. Blood chemistry levels were reported as follows: prothrombin time, 15.4 seconds; total bilirubin, 1.20 mg. per cent; direct bilirubin, 0.63 mg. per cent; cephalin flocculation test, negative in forty-eight hours; blood urea nitrogen, 16.9 mg. per cent; and uric acid, 4.4 mg. per cent. Results of indirect and direct Coombs' tests were negative. Although the stools were 4-plus guaiac-positive, a gastrointestinal x-ray series did not reveal any abnormalities. The bone marrow aspirate was normocellular and showed a slight eosinophilia. The urine did not reveal any abnormalities.

The patient was given a conservative ulcer regimen and supportive treatment. Prednisone was discontinued and blood transfusions were given. The first day after hospitalization a pericardial friction rub and gallop rhythm became evident, and a paradoxical pulse was present. An electrocardiogram did not show specific changes of pericarditis. Following the blood transfusions, no platelets were noted on peripheral smears. The stools remained guaiac-positive during the subsequent course, and on one occasion epistaxis appeared. Repeated blood transfusions were necessary in order to maintain the hemoglobin at adequate levels. ACTH was given without beneficial effect. The bilirubin rose to total 4.22 mg. per cent, with 3.08 mg. per cent direct fraction in the serum. Bile appeared in the urine and increased urobilinogen was present. Results of indirect and direct Coombs' tests remained negative. The cephalin flocculation test was between 1-plus and 2-plus in twenty-four hours; thymol turbidity, 0.80 units; prothrombin time, 15.5 seconds; alkaline phosphatase, 10.6 Bodansky units. The glutamic oxaloacetic transaminase ranged from 15 to 30 units per ml. and the glutamic pyruvic transaminase from 17 to 42 units per ml. The hemoglobin level was maintained with repeated blood transfusions throughout. Several thoracenteses yielded a straw-colored, and later, a bloody pleural fluid. Two units of platelet transfusions did not change the platelet count. The patient was also digitalized and given antibiotics. Radiotherapy to the mediastinum did not result in improvement.

During the last week of his hospital course a bulging mass in the left upper quadrant developed which seemed to arise from the inferior rib cage. The spleen became palpable. Therapy with intravenous triethylene melamine (TEM) did not alter the relentless course. Respiratory acidosis developed and the patient was given Diamox,® 750 mg. daily, and put into a Bennett respirator. The treatment did not alter his clinical situation, and he passed into stupor and died on the forty-second hospital day. During this hospitalization the patient received sixteen units of blood. His total duration of disease from the first symptom was twenty-seven months.

At autopsy the liver weighed 1,800 gm. At gross examination the right lobe was normal in appearance, and the capsule was smooth, homogeneous and brown. The left lobe, especially the dome, was infiltrated with firm, rubbery, gray tumor. On cut section the liver presented essentially the picture of congestion. The hepatic veins and arteries as well as the portal vein were patent. Microscopically (Fig. 4) Hodgkin's disease was present in both lobes of the liver. Central hemorrhagic necrosis, central vein sclerosis, and bile stasis were present.

The spleen weight was 350 gm.; the capsule was smooth. On section the spleen was soft and the surface was a homogeneous red in color. The tumor in the hilum of the spleen did not penetrate the capsule. The splenic vein was surrounded and partially compressed by tumor, the lumen being diminished to a caliber of approximately 0.5 cm. Microscopically, infiltrating Hodgkin's disease was seen in the hilum of the spleen, and congestion was present.

When examined macroscopically, the course of the esophagus was seen to be somewhat distorted by the presence of posterior mediastinal lymph nodes. At the point where the esophagus penetrated the disphragm the lumen was stenosed, measuring 1 cm. in diameter. Esophageal varices were present microscopically. (Fig. 5.)

Macroscopically the lesser curvature of the stomach

was deformed by the presence of large tumor nodules arising in the posterior zone of the lesser curvature. The mucosa over these nodules was intact but congested. Microscopically infiltration of Hodgkin's disease in the stomach was seen.

The heart weight was 310 gm. The pericardial cavity was opened with difficulty. The parietal pericardium was thickened and fibrous. Both the parietal and visceral pericardium were covered with a fibrinous, shaggy, hemorrhagic-like material. The valves were normal; the coronary arteries were thin, delicate and patent throughout. Microscopically, acute hemorrhagic fibrinous pericarditis was seen. Increased interstitial perivascular fibrosis was present in the myocardium.

The thoracic inlet was filled with fixed, firm, fibrous, rubbery tumor involving the superior portion of the upper lobe of the right lung. The weight of the right lung and tumor combined was 900 gm. The lower lobe of the left lung was involved by tumor as well. Its weight was 1,000 gm. At one point the tumor had perforated the diaphragm and extended into the liver. Hodgkin's disease also involved the diaphragm, posterior mediastinum, hilum of the spleen, stomach and the root of the neck. Also present were pleuritis, bronchopneumonia with micro- and macroabscesses, radiation fibrosis of the lung, left hydroureter, hypoplasia of the bone marrow and testicular atrophy.

FIG. 5. Case II. Esophagus. Varices filled with red blood cells.

COMMENTS

Esophageal varices are common in portal hypertension regardless of its etiology. Portal hypertension may result from: (1) venous block within the portal tree or liver, (2) impaired outflow of blood from the liver, and (3) excessive splanchnic blood flow or excessive hepatic arterial blood flow [*5*]. The discrepancy between the available venous bed and blood flow can be located in three areas: (a) in the liver itself (intrahepatic portal hypertension), (b) in the portal vein (infrahepatic portal hypertension), and (c) in the suprahepatic area (suprahepatic portal hypertension). Examples of the last variety are congestive heart failure, tricuspid insufficiency and constrictive pericarditis [*5*]. The most common cause of portal hypertension is the intrahepatic variety; and within this group, cirrhosis of the liver in all its forms is the main cause [*5–7*].

Esophageal varices have also been found in conjunction with other intrahepatic processes such as viral hepatitis, amebic hepatitis, portal fibrosis, fatty metamorphosis of the liver [*7,9,10*], gumma of the liver [*6,18*], echinococcus cysts of the liver [*8*], sarcoidosis of the liver [*11*], schistosomiasis of the liver [*5,18*], and in primary and metastatic liver tumors [*6–8,12–17*]. It should be stressed that the presence of portal hypertension has not been proved, by direct measurements, in viral and amebic hepatitis [*5*].

Obstruction of the hepatic vein or its branches within the liver on an inflammatory or other basis also results in intrahepatic portal hypertension [*5,18*]. On the other hand, esophageal varices have been noted in pregnancy and in persons in whom no disease of the liver or portal system was found at autopsy [*5,19–22*].

Esophageal Varices and Cancers. Primary cancer of the liver: The incidence of esophageal varices in primary cancer of the liver is high. In one series of two hundred cases, 127 patients or 63.5 per cent had varying degrees of varicosities in the lower esophagus. Of these cases, 89 per cent were associated with portal thrombi [*14*]. Another authority on the subject of primary cancer of the liver stated that esophageal varices are present in almost all cases of primary cancer of the liver at necropsy [*23*]. According to Ewing [*25*], 85 per cent of the hepatomas and 50 per cent of the biliary carcinomas are associated with cirrhosis. Fatal hemorrhages from esophageal varices in primary cancer of the liver have been reported [*12–14*]. The presence of esophageal varices in primary cancer of the liver can be understood in view of the fact that cirrhosis of the liver is present in a large percentage of these patients and that the tumor has a tendency to invade the portal vein branches and the tributaries of the hepatic vein as well.

Metastatic carcinoma of the liver: Eleven cases of esophageal varices in patients with metastatic carcinoma of the liver were found in the American literature [*7,15–17*]. In one patient [*7*] the metastatic nodules were small and not numerous; the cause of esophageal varices remains unclear in this case since it is difficult to explain their occurrence on mechanical embarrassment of the intrahepatic portal circulation. In the other cases, it seems that the varices were a result of portal flow diminution within the liver due to the metastases.

Lymphoma: In a recent review of 1,269 cases of lymphosarcoma with 250 postmortem examinations, no instance of esophageal varices was found [*26*]. To our knowledge, no case of esophageal varices has been reported in patients with leukemic involvement of the liver unless concomitant cirrhosis of the liver was present as well. In a study of 300 autopsies performed in children with leukemia no instance of esophageal varices was reported [*28*]. In a series of forty-four autopsies of leukemic adults* seen by the Chemotherapy Service of the Department of Medicine at Memorial Center, no instance of esophageal varices was seen [*29*].

In our first patient involvement of the liver with Hodgkin's disease was very extensive. Our second patient also had diffuse and extensive liver involvement with Hodgkin's disease; but in addition, a suprahepatic cause of portal hypertension was present, namely constrictive pericarditis. It is possible that the involvement of the thoracic inlet by Hodgkin's disease and the distortion of the esophagus caused by nodes may have obstructed the azygos vein and/or the esophageal venous drainage, and thus could have contributed to creation of the portal hypertension. In our two patients, neither cirrhosis of the liver nor changes in the portal or hepatic veins were seen.

In our opinion, more than one factor contributed to the development o the esophageal varices in our second patient, and it is difficult to assess the quantitative role played by each. It should be pointed out that measurements of portal pressure were not made in our patients, and therefore the presence of increased portal pressure is entertained on circumstantial evidence only. The jaundice in both cases is attributed to extensive liver involvement with disease, although an additional hemolytic factor cannot be ruled out since no red blood cell survival studies were made.

That the hemorrhage in the first case derived from the esophageal varices was proved at autopsy. It is probable that this event precipitated the hepatic coma. We believe that the bleeding source in the second patient was the varices since the lesion in the stomach was interstitial and the overlying mucosa was intact. The presence of thrombocytopenia in both cases probably was a contributing factor to the hemorrhage.

Another interesting finding in the first patient was the invasion of the gallbladder by Hodgkin's disease. This feature of the disease has been described previously [*24*].

* The series consists of twenty-nine cases of acute leukemia, seven cases of myelogenous leukemia in the acute phase, five cases of chronic myelogenous leukemia, and three cases of chronic lymphatic leukemia.

SUMMARY

Two cases of Hodgkin's disease with esophageal varices are reported. Both patients had extensive liver involvement with Hodgkin's disease, and the second patient had, in addition, constrictive pericarditis. To the best of our knowledge, no cases of esophageal varices in patients with Hodgkin's disease involving the liver have been reported.

The occurrence of varices in cancers of the liver and other pathological conditions is discussed.

Acknowledgment: We are greatly indebted to Dr. Stephen S. Sternberg of the Memorial Center, Pathology Department, for his review of the pathological material.

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Coexistent Hypercoagulability and Acute Hypofibrinogenemia in a Patient with Prostatic Carcinoma*

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JURGENS and Trautwein [*1*] published the first report of hypofibrinogenemia in prostatic carcinoma in 1930, and attributed it to impaired production of fibrinogen. A second example did not appear until the paper of Marder and his co-authors in 1949 [*2*]. The case report of Seale and her associates [*3*] soon followed and first mentioned fibrinolysis and intravascular clotting as possible causes.

However, it remained for Tagnon and his coworkers [*4–6*] to focus attention upon hypofibrinogenemia in prostatic carcinoma and to demonstrate fibrinolytic activity in such patients' blood. They also reported that proaccelerin (factor v, plasma Ac-globulin) and prothrombin levels fell. They concluded that proteolysis caused both the hypofibrinogenemia and the other clotting factor deficiencies, a conclusion subsequent authors have accepted [*7–10*].

Acute hypofibrinogenemia may also develop in other malignancies, in certain obstetrical complications and after surgical procedures. Much evidence favors intravascular clotting as the cause for hypofibrinogenemia in these circumstances.

The patient with prostatic carcinoma to be described had hypofibrinogenemia associated first with bleeding and then with unequivocal evidence of hypercoagulability. The simultaneous hypofibrinogenemia and hypercoagulability strongly suggests that intravascular clotting, rather than fibrinolysis, caused his fibrinogenopenia.

FIG. 1. Examples of the large ecchymoses that formed during the bleeding episode.

CASE REPORT

C. C., a sixty-six year old white man, first entered the Long Beach V. A. hospital (94853) on May 26, 1956, because of pain. Mild, aching pain in the back and hip had been present for about one year, and cramping epigastric pain for about five months. Nausea and vomiting had accompanied the latter and had led to a 30 pound weight loss. Pain in the anterior portion of the chest, aggravated by a cough productive of blood-tinged sputum, had appeared about two months prior to admission.

He gave no history of urinary tract symptoms, disease of the liver or unusual drug ingestion. Two teeth had been extracted several years earlier without unusual bleeding. No family history of bleeding was obtained.

Physical examination revealed a well developed, elderly white man with normal vital signs. Dried blood was seen in his left ear canal and around several alveolar sockets. A greatly enlarged, stony hard, nodular prostate was felt on rectal examination;

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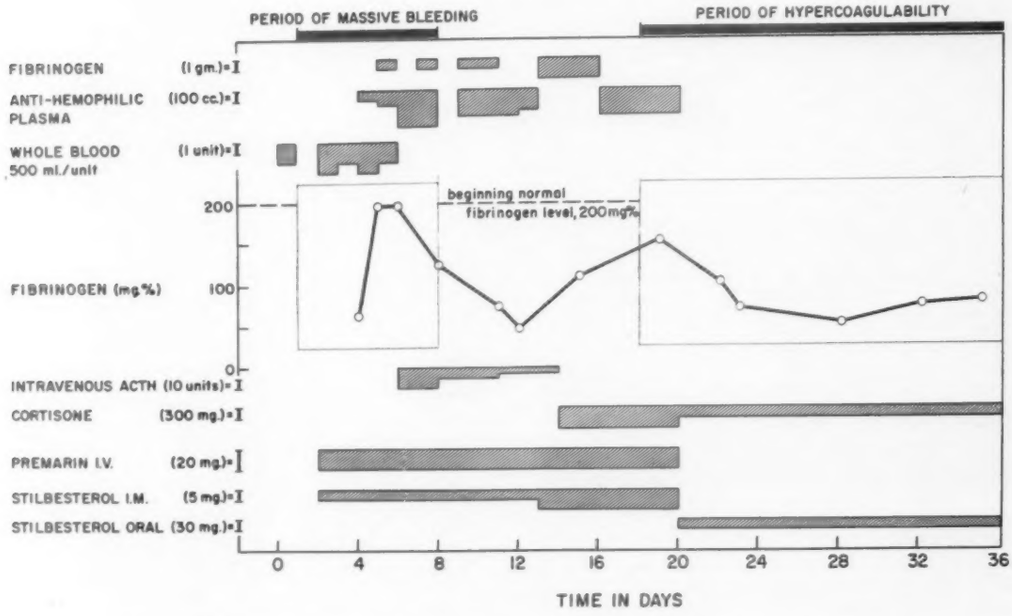

FIG. 2. Fibrinogen levels and therapy during the initial period of observation.

the borders of the prostate fused with the rectal wall. Blood was noted on the rectal glove. The remainder of his physical examination was within normal limits.

On admission the hemoglobin was 14 gm. per cent, hematocrit 43 per cent, white blood cell count 4,400 per cu. mm., platelet count 92,000 per cu. mm., and erythrocyte sedimentation rate (Wintrobe, uncorrected) 10 mm. Heavy proteinuria and masses of red blood cells were found on urinalysis. His blood urea nitrogen, serum proteins, bilirubin, cephalin flocculation and thymol turbidity tests were all within normal limits.

Cancer cells were seen on smears and sections of an iliac marrow aspirate. The serum acid and alkaline phosphatase values were 18.5 units and 17.6 Bodansky units, respectively. X-ray films of the bones were normal except for an osteolytic lesion of the left seventh rib.

Melena and microscopic hematuria were noticed during the patient's first three hospital days. On the fourth day he fainted while returning from the bathroom. His hemoglobin had fallen to 12.9 gm. per cent, his hematocrit to 35 per cent. A Lee-White clotting test was reported as showing no clotting in forty-five minutes.

By the next day, June 1, widespread bleeding was evident. As Figure 1 shows, large ecchymoses formed at injection sites and around bruises received during his fainting episode. Blood oozed for several hours from needle punctures. Evidence of retroperitoneal bleeding, gross hematuria, increased hemoptysis and hematemesis appeared. He was given large quantities of bank blood, reconstituted fresh frozen plasma* and fibrinogen.† He also received 20 mg. of Premarin® intravenously and 25 mg. of stilbestrol intramuscularly initially; thereafter, 20 mg. of intravenous Premarin and 5 mg. of intramuscular stilbestrol was administered daily. Intravenous ACTH therapy, 20 units daily, was started on June 6. Figure 2 summarizes the patient's therapy.

Bleeding did not definitely stop until June 8. On that day the urine and gastric contents were free of gross blood, needle puncture sites no longer oozed and the ecchymoses had ceased enlarging. His clinical condition gradually improved from then on. Nevertheless, fibrinogen levels on June 11 and 12 were only 70 and 47 mg. per cent, respectively. To forestall bleeding, plasma and/or fibrinogen was given through June 19. This raised his fibrinogen level to 160 mg. per cent, but it fell again when transfusions were discontinued. (Fig. 2.) As discussed later, striking evidence of hypercoagulability accompanied this second fall.

Premarin therapy, administered intravenously, was replaced on June 21 by 30 mg. of oral stilbestrol daily, and ACTH, administered intravenously, was replaced on June 14 by 600 mg. of oral cortisone daily. The cortisone was reduced to 300 mg. daily on June 20, and over the next few months to a maintenance level of 75 mg. daily. Hormone therapy resulted in a fall in serum acid phosphatase from 18.5 units on admission to 1.4 units on June 22, and in marked

* Irradiated antihemophilic plasma, Hyland Laboratories, Los Angeles, California.

† Parenogen,® Cutter Laboratories, Berkeley, California.

TABLE 1

SUMMARY OF COAGULATION STUDIES (NORMAL FIGURES IN PARENTHESES)

Date (1956)	Fibrinogen (200–400 mg. %)	Platelets (150,000–250,000 per cu. mm.)	Quick "Prothrombin" Time (100%)	Prothrombin Proconvertin (100%)	Proaccelerin Time (100%)	Venom Prothrombin Time (100%)	Partial Thromboplastin Time (50–90 sec.)	Thrombin Time (20–26 sec.)
5/29	...	92,000	.....	...	...	...	.....	.....
6/1	...	100,000	36	84	50	28	71, 71	92, 94
6/4	59	100,000	.....	...	...	33	.....	>180
6/6	194	84,000	95	95	50	45	64, 65	44, 50
6/8	125	.....	.....	...	...	...	.....	.....
6/11	72	.....	.....	...	...	...	.....	.....
6/12	47	170,000	.....	...	...	...	.....	.....
6/14	145	.....	.....	...	...	...	.....	.....
6/18	...	160,000	100	100	86	100	31, 32	.....
6/19	160	.....	.....	...	...	...	.....	.....
6/22	110	.....	.....	...	...	...	.....	.....
6/23	70	.....	.....	...	...	...	.....	.....
6/26	...	.....	70	130	300	95	67, 65	.....
6/28	57	100,000	.....	...	...	...	.....	.....
7/3	76	.....	.....	...	...	...	.....	.....
7/11	76	130,000	.....	100	96	100	56, 62	37, 36
7/20	41	130,000	.....	...	...	...	.....	.....
8/14	106	96,000	.....	...	...	...	.....	.....
9/5	...	170,000	140	200	140	100	45, 43	36, 37
9/12	...	.....	>150	200	72	85	47	31, 32
10/10	140	140,000	.....	...	...	...	.....	.....
10/21	...	.....	.....	...	...	...	.....	.....
10/25	225	150,000	.....	...	...	...	.....	.....
11/7	272	180,000	.....	...	...	...	.....	.....
12/18	290	272,000	.....	...	...	...	.....	.....
12/27	270	250,000	115	105	185	55	51, 49	17, 18

NOTE: 6/4/56 = Results of thromboplastin generation test, normal.
6/21/56 = Lee-White clotting time in glass: five and a half minutes, seven minutes, eight and a half minutes; in silicone coated tube, thirteen minutes (normal twenty to twenty-five minutes).
7/11/56 = Lee-White clotting time in glass: clotting in all three tubes, within five minutes; no clot lysis in twenty-four hours.
10/21/56 = Lee-White clotting time in silicone coated tube, thirteen minutes.

shrinking of the tumor felt on rectal examination. The patient did well for the next several months. However, the fibrinogen levels persisted between 40 and 140 mg. per cent until October, when they rose to normal. The serum acid phosphatase varied between 0.7 and 3.8 units. In mid-December it rose to 8.0 units.

The patient was readmitted to the hospital in January 1957, because of sudden, severe pain in the left side of the chest. Jaundice and findings of consolidation at the left lung base were evident. His left leg was swollen. A density in his lower left lung was seen on a chest roentgenogram; electrocardiographic findings suggested a pulmonary embolus. Liver function tests were compatible with hepatocellular disease.

The diagnoses of pulmonary embolism and serum hepatitis were made. Because of the latter, anticoagulants were not given. He appeared to be doing well until February 6, when he suddenly coughed up blood, went into shock and died.

On autopsy examination (performed by Dr. C. R. Payseur), an adenocarcinoma of the prostate was found, which had spread to the bladder, seminal vessicles, lumbar vertebrae and ribs. The liver appeared normal on gross examination but degeneration and necrosis compatible with serum hepatitis was seen on microscopic examination. A thrombus occluded the left femoral vein and extended into the left iliac vein. A large, partially organized embolus was found in a major branch of the left pulmonary artery. Smaller emboli were seen on microscopic examination in other pulmonary vessels. Areas of recent pulmonary infarction were found in both lungs.

COAGULATION STUDIES

Methods. Plasma was obtained by mixing nine parts of venous blood with one part of 2.0 per cent sodium oxalate solution. Our techniques

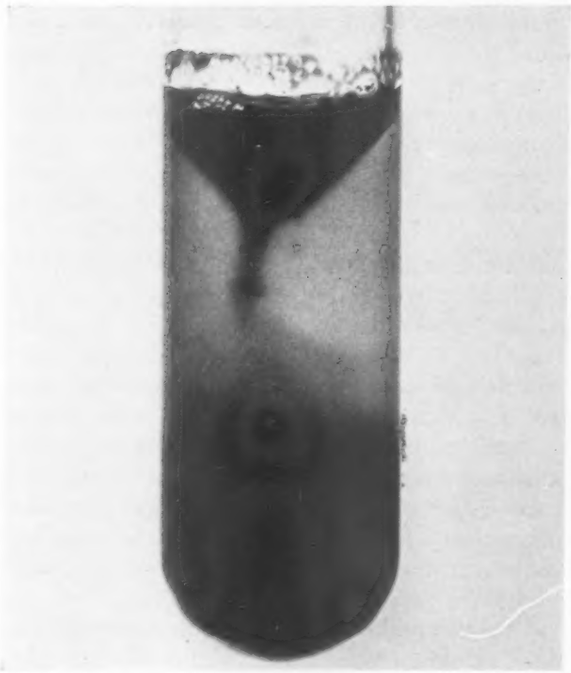

FIG. 3. The small clot of June 4, still present after standing twelve hours.

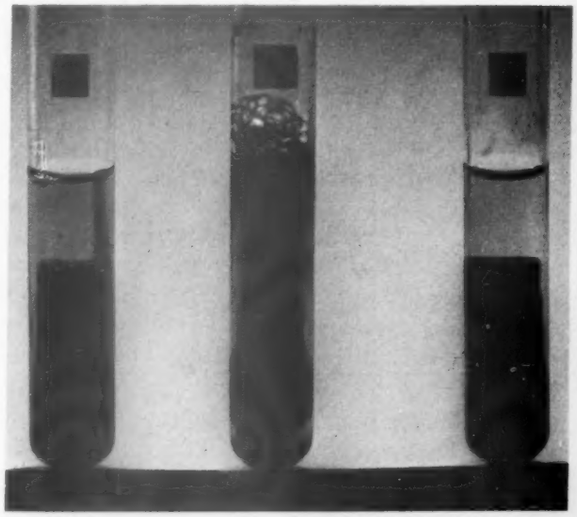

FIG. 4. The patient's blood clot (tube 1); a clot formed from a mixture of the patient's blood and normal blood (tube 2); and a normal blood clot (tube 3); all tubes after standing twenty-four hours at 37°C. Blood samples were drawn on June 13.

for the Lee-White clotting test, Quick one-stage "prothrombin" time, prothrombin-proconvertin test of Owren and Aas, one-stage proaccelerin assay, venom-"cephalin" prothrombin assay, partial thromboplastin time and plasma thrombin time have been published elsewhere [*11*]. The turbidometric method [*12*] was used to measure fibrinogen (normal 200 to 400 mg. per cent) and the Rees-Ecker method to count platelets (normal 150,000 to 300,000 per cu. mm.). The results of fibrinogen, platelets and the clotting tests over a seven-month period are listed in Table I.

Results of Tests for Fibrinolysis. Repeated search yielded only very questionable evidence of *in vitro* fibrinolysis. Blood in a Lee-White test performed on May 30 failed to clot, presumably because of afibrinogenemia. Conceivably, a small clot could have formed and lysed without being noticed. On June 1, the patient's blood formed a very small clot which disintegrated within three hours, either due to its inherent instability or to *in vitro* fibrinolytic activity. Blood taken on June 4 again formed a small clot which was still present twelve hours later. (Fig. 3.) We attempted to demonstrate fibrinolysis on June 13, after the patient had received enough fibrinogen to make a good clot. Figure 4 shows this clot (tube 1), a clot made up of equal parts of his and of normal blood (tube 2), and a normal blood clot (tube 3) after standing for twenty-four hours at 37°C. All are intact. Moreover, his plasma fibrinogen values were identical before and after standing for twenty-four hours at 37°C. Despite the persistence of hypofibrinogenemia, whole blood clots on July 7 and 11 also failed to lyse in twenty-four hours.

Thrombin Times. Very long thrombin times were found during the bleeding episode. These were not due to a quantitative fibrinogen deficiency alone. A fibrinogen level of about 100 mg. per cent is reportedly optimal for the thrombin-fibrinogen reaction [*13*]. The patient's thrombin time on June 6 was about twice normal despite a fibrinogen level above 125 mg. per cent.

Ratnoff and Vosburgh [*14*] demonstrated thrombin-inhibitory activity in the plasma of a patient with hypofibrinogenemia complicating amniotic fluid embolism. Table II lists the thrombin times of mixtures of our patient's plasma of June 1 and normal plasma. These are hard to interpret. Thrombin times were shortened when the ratio of normal plasma to patient's plasma was 1:2, and completely corrected when it was 2:1. This argues against thrombin-inhibitory activity in the patient's plasma. In contrast, the slightly prolonged time of the 1:1 mixture suggests that mild thrombin-inhibitory activity did exist.

Prothrombin, Proaccelerin and Proconvertin During the Bleeding Episode. A Quick one-stage prothrombin time was 36 per cent on June 1, and

TABLE II
THROMBIN TIMES ON JUNE 1 OF MIXTURES OF PATIENT'S PLASMA AND NORMAL PLASMA

Patient's Plasma (parts)	Normal Plasma (parts)	Thrombin Time (normal, 20–26 sec.)
1	0	92, 95
2	1	36
1	1	31
1	2	26

95 per cent on June 6. Prothrombin, estimated by the more specific Russell's viper venom-cephalin assay of Hjort and his associates [*15*], was 28, 33 and 45 per cent on June 1, 4 and 6, respectively. Proaccelerin levels were 50 per cent of normal during the bleeding period. In contrast, almost normal values were obtained in the prothrombin-proconvertin assay of Owren and Aas [*16*], indicating that proconvertin (factor VII, stable factor) activity was not depressed. Thus prothrombin deficiency appeared to be the major deficiency during the bleeding phase, with a moderate accompanying proaccelerin deficiency and with no evidence of proconvertin deficiency.

Plasma Thromboplastic Factors During the Bleeding Episode. Partial thromboplastin times on June 1 and 6 were within our normal range of fifty to ninety seconds. Results of a thromboplastin generation test performed on June 4 were normal. Thus we could not demonstrate an antihemophilic factor (antihemophilic globulin, plasma thromboplastin precursor and plasma thromboplastin antecedent) deficiency during the bleeding episode.

Platelet Counts. Mild thrombocytopenia persisted throughout both the bleeding episode and the period of hypofibrinogenemia associated with hypercoagulability. The platelet count and the fibrinogen level rose to normal together.

Evidence of Hypercoagulability. On June 18, ten days after active bleeding had stopped, a partial thromboplastin time of thirty-one seconds was noted. This value, almost twenty seconds below normal, is the shortest partial thromboplastin time in four years' experience in this laboratory. Three days later, on June 21, the clinical laboratory was unable to obtain plasma for chemical studies because the patient's blood clotted despite rapid mixing with a dried oxalate anticoagulant. On June 26 we found a small clot in blood taken for coagulation studies in liquid oxalate anticoagulant. The prothrombin-proconvertin time of this "plasma" was 130 per cent, suggestive of proconvertin activation during clotting [*17*]. Its proaccelerin level was more than 300 per cent of normal, an indication that thrombin had converted part of the proaccelerin (plasma Ac-globulin) to accelerin (serum Ac-globulin) [*18*].

Clotting in oxalate anticoagulant was demonstrated again on June 27. Blood was drawn rapidly into a silicone-coated syringe through a No. 19 Arquard-coated needle. Nine ml. were added without delay to 1 ml. of 2 per cent liquid sodium oxalate, and 10 ml. to dried oxalate anticoagulant. Both tubes were inverted and mixed ten times. A clot did not form immediately, but after ten minutes a small clot was noticed in both tubes. It disappeared after four hours.

Hypercoagulability was also demonstrated on July 11, when Lee-White clotting times in glass were under five minutes in all three tubes; on September 5, when the Quick, prothrombin-proconvertin, proaccelerin, and partial thromboplastin time tests were all rapid; and on September 12, when the Quick, prothrombin-proconvertin, and partial thromboplastin time tests were again faster than normal. Suggestive evidence of hypercoagulability was still present on October 21, when blood clotted in a silicone-coated tube in thirteen minutes, and on December 27 when the Quick and proaccelerin values were still elevated.

We emphasize that the striking evidence of hypercoagulability was obtained during a period of progressive hypofibrinogenemia. (Fig. 2, Table I.) Thus the patient's fibrinogen level, boosted by transfusions to 160 mg. per cent on June 19, fell to 57 mg. per cent by June 29. During this period, on June 21, 26 and 27, his blood clotted *in vitro* despite rapid mixing with oxalate anticoagulant.

COMMENTS

The complete syndrome of acute hypofibrinogenemia consists of (1) fibrinogenopenia, (2) diminished proaccelerin, prothrombin and antihemophilic globulin levels, (3) moderate thrombocytopenia, and (4) increased fibrinolytic activity. Every patient will not show all of these defects. In particular, patients need not have evidence of *in vitro* fibrinolysis [*8,19–24*]. Throm-

bocytopenia may not occur. Proaccelerin deficiency has been repeatedly reported and undoubtedly contributes to the lengthening of the Quick prothrombin time that is almost always found. True prothrombin deficiency, measured by specific methods, has also been reported [*20,25,26*] but not often enough to evaluate its frequency. Three recent studies indicate that antihemophilic globulin levels also fall [*20,27,28*]. Despite these multiple clotting factor defects the hypofibrinogenemia probably causes the bleeding, since the intravenous injection of enough concentrated fibrinogen usually stops it.

Two theories explain the fibrinogen destruction that leads to the fibrinogenopenia. Both assume that tissue extracts enter the blood. One states that such extracts act primarily as thromboplastins. They initiate widespread intravascular clotting that consumes fibrinogen and other clotting factors faster than they are made. Fibrinolysis may also occur and aggravate the deficiencies, but it is considered a secondary, perhaps compensatory, response aimed at dissolving fibrin plugs.

The second theory states that fibrinolysis is the primary event. Its advocates believe that circulating tissue extracts, either directly or by activating plasmin, can lyse enough fibrinogen and other clotting factors to produce the syndrome. A third, combined opinion has also been offered [*20*], namely, that circulating tissue extracts induce both processes, acting as thromboplastins in low concentration and fibrinolytic enzymes in high concentration.

An extensive literature supports the view that intravascular clotting initiates the acute hypofibrinogenemia syndrome. It may be summarized as follows:

(1) *Intravascular clotting best explains the multiple clotting factor defects found in the acute hypofibrinogenemia syndrome:* The slow intravenous injection of thromboplastin into animals produces clotting factor deficiencies closely resembling those described in acute hypofibrinogenemia. Thus Mellanby discovered in 1909 [*29*] that the intravenous injection of thromboplastic material into animal depletes the blood of fibrinogen. Many others have confirmed this with a variety of thromboplastic agents [*30–35*]. Hartmann and his co-workers found [*34*] and Penick and his associates have confirmed [*36*] the finding that dogs injected with thromboplastin also become thrombocytopenic. Mustard has stated [*37*] that humans become thrombocytopenic when transfused with blood containing thromboplastic material formed as bank blood stands. Moreover, antihemophilic globulin levels fall when dogs are injected with thromboplastin [*36*].

The Quick prothrombin time lengthens after intravenous thromboplastin injection [*27,34*]. Specific assay techniques have revealed both prothrombin and proaccelerin deficiencies [*25*]. The thrombin time also lengthens, not entirely because of reduced fibrinogen levels [*33,34*].

Thus one finds diminished fibrinogen, platelets, antihemophilic globulin, prothrombin and proaccelerin both in animals injected with thromboplastin and in patients with acute hypofibrinogenemia. Moreover, Frick has pointed out [*20*] that the same pattern develops when normal blood clots spontaneously *in vitro*.

Could fibrinolysis explain these multiple defects? Fibrinolysins can destroy proaccelerin [*6,38–41*]. However, thrombocytopenia due to fibrinolytic activity has not been demonstrated experimentally. An early report that fibrinolysin destroys prothrombin [*42*] has been refuted [*38*] and probably represents another example of proaccelerin inactivation. Indeed, prothrombin seems very resistant to fibrinolytic activity [*39,40*]. Finally, it is hard to believe that *in vivo* fibrinolysis could produce serious, multiple clotting factor defects without *in vitro* evidence of fibrinolysis.

Our clear-cut demonstration of simultaneous hypercoagulability and hypofibrinogenemia completes the evidence from coagulation studies supporting the intravascular clotting theory. We found one convincing previous report of hypercoagulability, that of Soulier and his co-workers [*43*] who described progressive shortening of the whole blood clotting time preceding fibrinolysis in patients undergoing pulmonary surgery. Graham and his associates [*44*] also reported that plasma from a patient with acute postoperative hypofibrinogenemia shortened the clotting time of normal blood, but these investigators did not study the effect of normal plasma treated similarly.

(2) *Intravascular fibrin deposits have been seen in a small but significant number of cases of acute hypofibrinogenemia:* Seale cited [*3*] an unreported case that she and Dr. Paul Aggeler studied in which the patient had prostatic carcinoma, uncontrollable bleeding, very low fibrinogen values and postmortem evidence of intravascular fibrin clots in several organs. Mckay and his co-workers

[45] described a patient with carcinoma of the pancreas who died of hypofibrinogenemic bleeding, but autopsy examination disclosed multiple and widespread intravascular clotting. These authors also mention McKay's unpublished observations of intravascular fibrin deposits in patients with premature separation of the placenta at autopsy. Tuller [46] has described the occurrence of amniotic fluid embolism, afibrinogenemia and disseminated fibrin thrombosis. Finally, Graham and his co-workers [44] have reported the striking case of a patient who became fibrinogenopenic and bled after hysterectomy, but who died with widespread intravascular thrombosis after fibrinogen administration. One can hardly doubt that intravascular clotting also initiated the hypofibrinogenemia in this patient.

(3) *Potent tissue thromboplastin could enter the circulation in those conditions in which acute hypofibrinogenemia occurs:* The obstetrical complications associated with acute hypofibrinogenemia are abruptio placenta, amniotic fluid embolism, retained dead fetus and septic abortion. In each, placental or amniotic material could gain access to the maternal circulation [23,25,47–51]. Placental extracts possess well established strong thromboplastic activity [32,52,53], and amniotic fluid may possess weaker thromboplastic activity [54].

Acute hypofibrinogenemia has been reported as a complication of pulmonary resection [43,55]. Lung tissue contains a well known powerful thromboplastic activity that probably enters the circulation in varying amounts during the manipulations involved in resection.

Little is known about the thromboplastic properties of tumors or their products. However, acute hypofibrinogenemia has occurred in at least two patients with carcinoma of the body or tail of the pancreas [20,45]. Tumor in this location is associated with a notoriously high incidence of multiple thromboembolic lesions [56,57], strongly suggestive of coagulant material entering the blood [58].

Despite this evidence, proteolysis of fibrin and fibrinogen has usually been blamed for the acute hypofibrinogenemia complicating prostatic carcinoma. This stems from the emphasis Tagnon and his associates [4–6] placed upon fibrinolysis in this disease. Huggins and Neal [59] had shown that human prostatic fluid possesses fibrinolytic activity. Tagnon's group found similar activity in the blood of some patients with prostatic carcinoma and in extracts of prostatic carcinoma. Therefore, they postulated that prostatic carcinoma tissue could release a powerful fibrinolysin into the blood, capable of causing hypofibrinogenemia and hemorrhage.

Only Frick has seriously questioned this explanation [20]. Yet, several patients with acute hypofibrinogenemia complicating prostatic carcinoma have been described [8,19,20] without convincing evidence of *in vitro* fibrinolysis. Moreover, Tagnon defined fibrinolysis as clot liquefaction within twenty-four hours. This degree of fibrinolytic activity can be found without hypofibrinogenemia. For example, we have seen several patients with chronic disease of the liver whose fibrinogen levels were adequate to produce good clots, but whose clots dissolved on standing overnight. Cliffton's data [60] indicate that human subjects may be given enough plasmin intravenously to produce *in vitro* clot lysis within four hours, without bleeding or marked fibrinogenopenia.

Aggeler and Seale's unpublished case of multiple fibrin thrombi has already been referred to. We have discussed this patient with Dr. Aggeler and feel certain that this case represents an example of acute hypofibrinogenemia in prostatic carcinoma, induced by intravascular clotting.

Our patient's coagulation studies strongly suggest that intravascular clotting caused his hypofibrinogenemia. Evidence for *in vitro* fibrinolysis was most unconvincing. In contrast, hypercoagulability was unequivocally demonstrated. Although transfusions corrected his initial hypofibrinogenemia, it reappeared during the very period that samples of his oxalated blood would clot. Human prostatic fluid cannot clot oxalated plasma [61]. Therefore, this clotting must be attributed to thrombin formed from reactions started by circulating thromboplastic material. A recent description of oxalated blood from a dog which clotted after injection of thromboplastin supports this conclusion [35].

Our patient's course can be reconstructed as follows: His very high initial serum acid phosphatase proves that prostatic tumor products were entering his blood in quantity. This included enough thromboplastic material to produce diffuse intravascular clotting and resultant hypofibrinogenemia. His low prothrombin levels were one indicator, his moderate proaccelerin deficiency another [26], of a rapid rate of

intravascular clotting. His widespread bleeding resulted from a combination of fibrin-damaged blood vessels and of hypofibrinogenemia.

As the fall in his serum acid phosphatase illustrates, estrogen administration slowed the discharge of tumor products into his circulation. Intravascular clotting was reduced to a critical rate at which it was possible to demonstrate marked hypercoagulability [*35*]. This rate was fast enough for hypofibrinogenemia and thrombocytopenia to continue, but not fast enough to prevent his prothrombin level from returning to normal.

Intravascular clotting further slowed with further control of his disease. As a consequence, fibrinogen and platelets returned to normal and oxalated blood samples would no longer clot. Some evidence of increased activity of prothrombin and its accelerators persisted, perhaps also reflecting an effect of estrogen [*62*] or adrenal steroid [*63*] administration.

The presence of homologous serum hepatitis complicates the interpretation of his final clotting studies. The rise in his serum acid phosphatase level suggests that his tumor was escaping from estrogen control; the terminal thromboembolic episodes indicate that his hypercoagulability had increased.

The full-blown syndrome of acute hypofibrinogenemia appears to be an extreme manifestation of circulating thromboplastic material. The pattern Goodman described in carcinoma of the pancreas [*64*]—venous thrombosis resistant to coumarin anticoagulants but, because of increased prothrombin utilization, associated with paradoxical coumarin hypersensitivity—seems to us to be a less extreme but closely related manifestation of the same disturbance. Moreover, the well known occurrence of venous thrombosis in a variety of visceral carcinomas may be the most frequent but only the mildest manifestation of circulating thromboplastic material in malignancy.

We do not know what significance to attach to fibrinolysis in acute hypofibrinogenemia, except to emphasize that both the bleeding and the multiple clotting factor defects can occur in its absence. Fibrinolysis undoubtedly minimizes damage due to diffuse intravascular thrombosis. In so doing, it could aggravate hemorrhage. Fibrinolysis would seem to be a particular hazard when surgical procedures leave large raw surfaces. In this circumstance, minor increases in fibrinolytic activity, insufficient to produce hypofibrinogenemia, could conceivably lead to persistent oozing.

SUMMARY

Acute hypofibrinogenemia, associated first with bleeding and then with hypercoagulability, developed in a patient with metastatic prostatic carcinoma. The hypercoagulability strongly suggests that intravascular clotting, rather than fibrinolysis, caused his acute hypofibrinogenemia. This case is thought to link the rare syndrome of acute hypofibrinogenemia with the common observation of venous thrombosis in malignancy.

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Heatstroke and Jaundice*

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HEATSTROKE is an uncommon but serious problem which must be treated vigorously if the patient is to survive. The complications of heatstroke are many, but jaundice is a rarely recorded manifestation. In an analysis of 125 fatal cases of heatstroke, Malamud, Haymaker and Custer [*1*] had four patients in whom jaundice was clinically apparent and two additional patients with elevated icterus indices; twelve patients, however, had moderate to severe centrolobular hepatic necrosis. Austin and Berry [*2*], in an analysis of 100 cases of heatstroke in which there were seventeen deaths, do not mention jaundice as such but they note that 33 per cent of fifteen van den Bergh determinations made on the 100 patients were greater than 1.0 mg. per 100 ml., and 100 per cent of eleven determinations of prothrombin times were less than 50 per cent of normal. The pathological status of the liver in the seventeen fatal cases in this series is not mentioned. It seems appropriate, therefore, to report a case of heatstroke in which the patient recovered, and in which jaundice was a marked feature.

TABLE 1
TEMPERATURES AND BLOOD PRESSURES DURING PERIOD OF HYPERTHERMIA

Date	Time	Temperature (°F. rectal)	Blood Pressure (mm. Hg)
July 6	2:00 P.M.	107.8	120/80
	.....	.....	76/0
	3:20 P.M.	107.8	124/30
	4:00 P.M.	106.4	100/30
	4:45 P.M.	105.0	.....
	5:15 P.M.	103.4	.....
	5:30 P.M.	101.0	.....
	5:45 P.M.	.....	140/60
	7:30 P.M.	.....	70/40
	8:00 P.M.	102.0	.....
	8:30 P.M.	103.0	.....
	10:30 P.M.	102.4	.....
	11:15 P.M.	102.0	100/60
	12:00 P.M.	102.0	100/60
July 7	5:00 A.M.	101.0	.....
	8:30 A.M.	99.0	.....

CASE REPORT

A twenty-three year old white man, with approximately one month of military service, was undergoing basic training in Texas. Just before noon on July 6, while on the rifle range, he became nauseated, and shortly thereafter noted a cessation of sweating. On return to his barracks he felt weak, complained of dizziness and faintness, and at 1:30 P.M. he became unconscious. On admission to the U. S. Army Hospital, Fort Hood, at 2:00 P.M. he was comatose. He had Cheyne-Stokes respirations, his skin was hot, dry and pale; his rectal temperature was 107.8°F., pulse 180, and blood pressure, 120/80 mm. Hg. There were multiple purpuric spots on his extremities and he had a right subconjunctival hemorrhage. His eyes were fixed and deviated to the right; the pupils were miotic and unresponsive to light. No deep tendon reflexes or Babinski signs were elicited initially. The physical examination was otherwise within normal limits. During the examination his blood pressure dropped to 76/0 mm. Hg and his pulse rate was 160 to 180. Immediate treatment consisted of the application of iced towels and sponging with alcohol. Intravenous administration of 500 ml. isotonic saline solution with 100 mg. of hydrocortisone was begun, followed by 700 ml. of isotonic saline solution with 4 ml. of Levophed.® An airway was inserted and nasal oxygen was given. The patient was put in ice water and cooled, and temperatures and blood pressures at various times were noted. (Table 1.)

At 5:00 P.M. 50 mg. of Thorazine® was given and a half hour later an intravenous infusion of 1,000 ml. of saline solution containing 100 mg. of hydrocortisone and Levophed was begun, and the patient was taken to the ward. At 5:45 P.M. his extremities became spastic; hyperactive deep tendon reflexes were present; his pulse rate was 140, and he had a generalized convulsion. He was given 15 ml. of paraldehyde intramuscularly. His extremities became rigid, as seen in decerebrate rigidity. His respirations were shallow, with gasping and grunting. Oxygen was continued

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TABLE II

SUMMARY OF LABORATORY STUDIES

Date	White Blood Cells (per cu. mm.)	Polymorphonuclears (%)	Hemoglobin (gm./100 ml.)	Total Serum Bilirubin (mg./100 ml.)	Direct Bilirubin (mg./100 ml.)	Cephalin Flocculation	Thymol Turbidity (units)	Alkaline Phosphatase (units)	Bromsulphalein (% retention)	Prothrombin Time (% normal)	Urine Urobilinogen (units)	SGO Transaminase (units)	Total Protein (gm./100 ml.)	Serum Albumin (gr./100 ml.)	Serum Globulin (gr./100 ml.)
July															
6	17,150	80	13.5	....	...	.....	...	....	..	...	....	.....	...	...	...
7	.....	..	....	....	...	.....	...	.....	..	...	....	.....	...	...	...
8	19,050	97	13.5	....	...	.....	...	....	...	...	....	.....	...	...	...
9	.....	..	....	23.7	9.1	.....	...	....	..	...	....	.....	...	...	...
10	.....	..	....	....	...	.....	...	....	..	...	....	.....	...	...	...
11	6,700	..	12.0	....	...	.....	...	....	..	...	....	.....	...	...	...
12	.....	..	....	19.2	7.9	.....	...	....	..	...	0.13	131	...	...	...
15	.....	..	....	22.3	9.3	Negative	4.4	3.0	..	100	....	42.5	...	...	...
17	7,150	..	16.0	....	...	.....	...	....	..	...	....	.....	...	...	...
18	.....	..	....	14.2	5.7	2+	6.2	....	..	...	1.2	32.5	7.0	3.6	3.4
24	.....	..	....	5.4	2.4	Negative	3.7	3.8	..	...	0.63	27.0	...	...	...
Aug.															
1	.....	..	....	3.9	...	Negative	4.0	4.2	..	...	0.59	27.5	...	...	...
8	.....	..	....	2.9	1.0	2+	3.0	....	..	...	0.2	.....	...	...	...
15	.....	..	....	1.2	0.0	.....	3.7	....	..	...	0.48	.....	...	...	...
Sept.															
14	.....	..	....	....	...	2+	...	....	..	...	....	.....	...	...	...
19	4,750	67	14.5	....	...	.....	...	....	..	...	....	.....	...	...	...
23	.....	..	....	0.6	0.0	.....	4.7	2.8*	..	...	0.28	.....	...	...	...
Oct.															
18	5,600	62	15.8	....	...	.....	...	....	10	...	....	.....	...	...	...
22	.....	..	....	....	...	.....	...	....	..	...	....	50	...	...	...
28	.....	..	....	....	...	.....	...	....	..	70	....	.....	...	...	...
Nov.															
12	.....	..	....	....	...	.....	...	....	..	82	....	.....	...	...	...
16	8,900	62	16.9	....	...	.....	...	....	..	...	....	24	...	...	...
26	6,200	64	15.7	....	..	.....	...	....	..	...	....	.....	8.2	5.3	2.9
29	.....	..	....	0.2	0.2	1+	2.0	5.4†	..	...	....	.....	...	...	...
30	.....	..	....	....	...	.....	...	....	..	...	....	.....	...	...	...
Dec.															
2	.....	..	....	....	...	.....	...	4.0	..	...	....	.....	...	...	...
3	.....	..	....	....	...	.....	...	...	4	...	....	.....	...	...	...
9	5,600	56	15.7	....	...	.....	...	3.6	..	...	....	.....	...	...	...
16	5,900	71	15.7	....	...	.....	...	4.0	..	...	....	.....	...	...	...

ADDITIONAL DATA: Reticulocyte count on July 17, 0.7 per cent; platelet count on November 12, 124,000; fecal urobilinogen on July 17, 166 Ehrlich units per 100 gm.

* SJR units.

† Bodansky units.

and his airway was kept clear with suctioning. At 6:15 P.M. another convulsion occurred and he was again given paraldehyde (15 ml.). At 7:00 P.M., because of restlessness, another 50 mg. of Thorazine was given. At 7:30 P.M. his blood pressure was 70/40 mm. Hg, and 2 ml. of Levophed was added to his intravenous fluids. His gag reflex appeared at 8:00 P.M. At 8:30 P.M. he had a pulsus paradoxicus of 33 mm. Hg. His lungs remained clear. At midnight his blood pressure was 100/60 mm. Hg and remained stable thereafter. At 1:00 A.M. the rigidity was still present and at 2:25 A.M. his deep tendon reflexes were again obtainable and a bilateral Babinski sign was present. By 5:00 A.M. his rigidity had decreased somewhat, he was restless, and intermittently waved his extremities. His urinary output remained good throughout his hospital course. At 8:30 A.M. his temperature was 99°F. and remained normal thereafter. The administration of Levophed was discontinued, but 100 mg. of hydrocortisone was given intravenously. He became responsive to painful stimuli at this time. At 12:30 P.M. the Babinski sign disappeared. At 9:30 P.M. he spoke for the first time. Oral administration of fluids was begun at this time, and nasal oxygen was discontinued. By 7:00 A.M. on July 8 he responded slowly to questions and by 1:00 P.M. he was able to carry on a full conversation, to act appropriately, and he had complete recall of events except for the period of unconsciousness. On the morning of July 9 he was rational but weak and nauseated. He was still receiving fluids intravenously. At this time scleral icterus was noted.

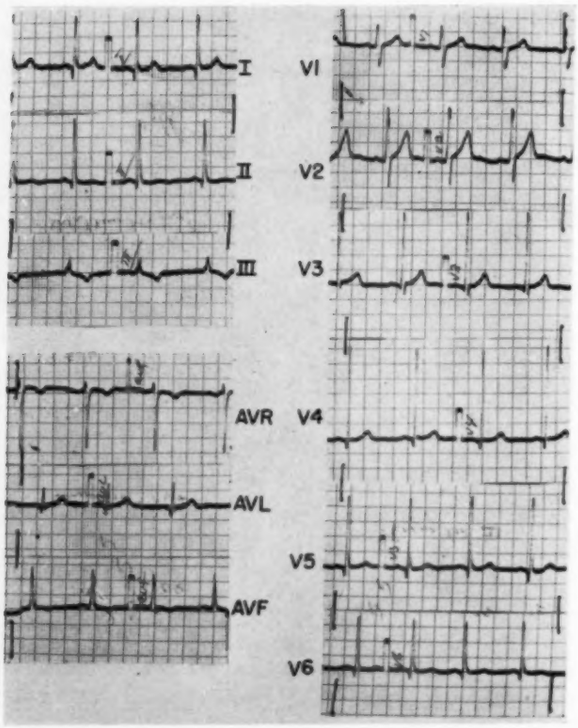

FIG. 1. Electrocardiogram taken on July 16, 1957.

Laboratory studies on admission showed a white blood count of 17,000 per cu. mm. with 80 per cent neutrophils. He had 4-plus glycosuria and 1-plus albuminuria; there were 3 to 6 red blood cells in his urine per high power field and 1 to 4 white blood cells and 1 to 4 granular casts, but these disappeared by the second hospital day. The blood sugar was 360 mg. per 100 ml. of blood but this was obtained while glucose was being given intravenously. On July 7 the serum sodium was 143 mEq. per L.; potassium, 3.7 mEq. per L.; chloride, 103 mEq. per L.; carbon dioxide, 36 vol. per cent; and on July 8, sodium was 137; potassium, 3.8; chloride, 107 mEq. per L. The white blood count was 19,050 per cu. mm. with 97 per cent neutrophils, the hemoglobin was 13.5 Gm. per cent. The serum electrolytes remained normal throughout his hospital course. On July 9 the total serum bilirubin was 23.7 mg. per 100 ml. of blood with a direct bilirubin of 9.1 mg. per 100 ml. of blood. By July 11 his white blood count was 6,700 per cu. mm. and remained normal thereafter. On July 12 the total serum bilirubin was 19.2 mg. per 100 ml. of blood with a direct bilirubin of 7.9 mg. per 100 ml. of blood. The serum transaminase was 131 units. On July 15 the serum transaminase was 42.5 units while the total serum bilirubin was 22.3 mg. per 100 ml. of blood, with a direct bilirubin of 9.3 mg. per 100 ml. of blood; thymol turbidity, 4.4 Maclaughlin units; alkaline phosphatase, 3.0 units; cephalin flocculation test, negative; prothrombin time, 100 per cent; cholesterol, 164 mg. per 100 ml. of blood with cholesterol esters of 80 mg. per 100 ml. of blood. On July 17 the fecal urobilinogen was 166 Ehrlich units per 100 gm.; hemoglobin, 16.0 gm. per cent; white blood count, 7,150 per cu. mm.; reticulocyte count, 0.7 per cent. By August 15 his total serum bilirubin was 1.2 mg. per 100 ml. of blood. The alkaline phosphatase, thymol turbidity and cephalin flocculation remained normal, and the serum transaminase had become normal. Table II shows the summary of laboratory studies.

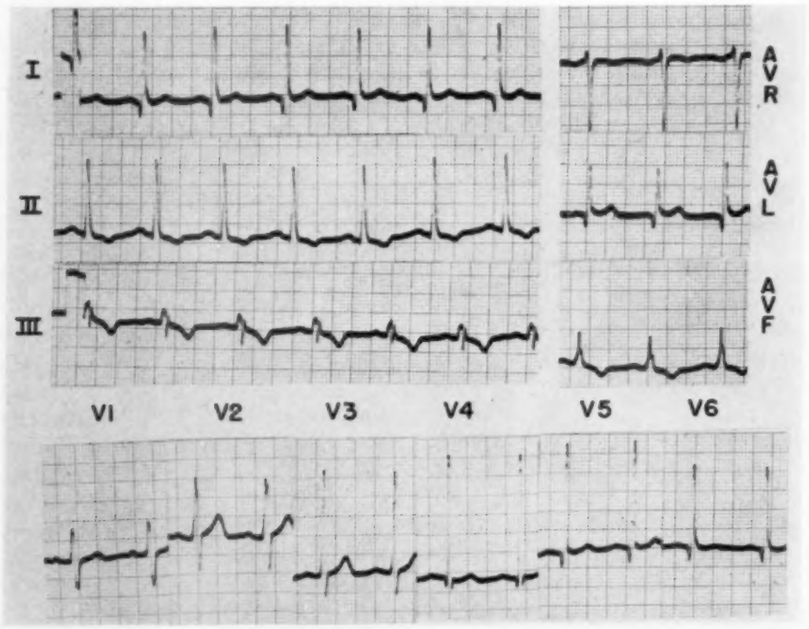

FIG. 2. Electrocardiogram taken on October 23, 1957.

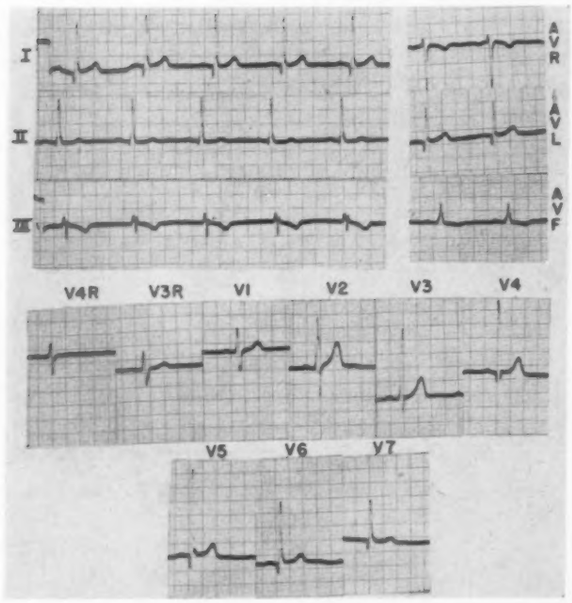

FIG. 3. Electrocardiogram taken on November 4, 1957.

During the next month the patient had changing neurological signs, including a positive Romberg sign, abnormal reflexes, and Parkinsonian-like movements. But by August 15 these had disappeared. He walked unsteadily at first and sometimes trembled. An electroencephalogram on July 22 was interpreted as being normal. He spent convalescent leave from August 20 until September 19 at home in Virginia. He stayed in bed most of the time and complained frequently of persistent frontal headache and stiffness and aching in both legs. The headache and aching in his legs persisted after he returned to the hospital and he limped on his right leg. In addition, lightheadedness and aching in his arms occurred intermittently, and he complained of swelling of his right leg. Often he could not sleep because of the persistent headache. No evidence of bone or joint disease was found by an orthopedic consultant, nor could any objective evidence of neurological or psychiatric disease be found. Because of these complaints he was admitted to the Walter Reed Army Hospital on October 16, 1957.

Physical examination at that time revealed a well developed, moderately obese white man who limped. He was 5 feet 6 inches tall and weighed 174 pounds. The pulse was 96, the blood pressure was 134/80 mm. Hg. The liver and spleen were not palpable. He had no icterus, spider nevi or palmar erythema. He complained of aching in his legs and burning headache. Neurological examination was within normal limits except for decreased vibratory sense below the right knee with some weakness in dorsiflexion of the right foot and hyperactive reflexes of both lower extremities. Otherwise the examination was within normal limits.

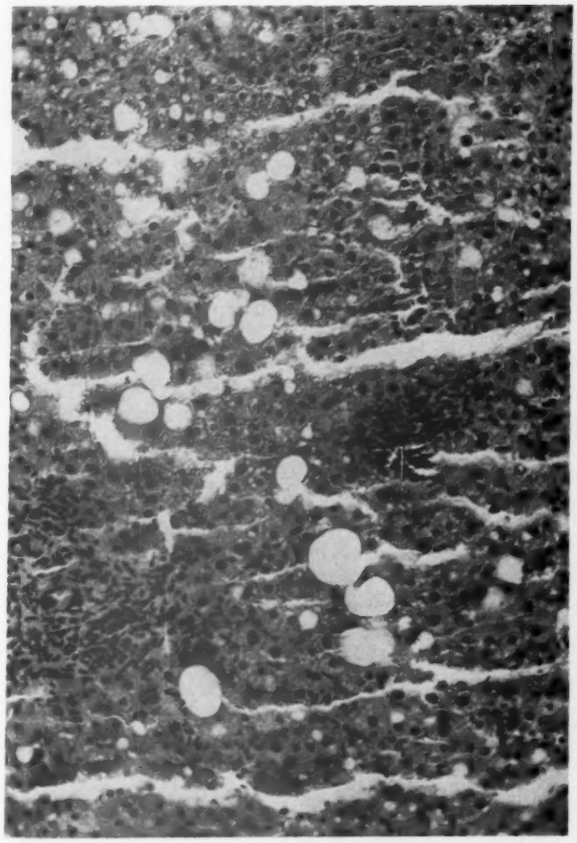

FIG. 4. Liver biopsy specimen showing fatty metamorphosis and focal infiltrate of lymphocytes.

Laboratory studies are included in Table II. His bromsulphalein (BSP) retention was 10 per cent on October 18, but was 4 per cent on December 3. The serum transaminase on October 22 was 50 units (normal, 10 to 40 units) but was 24 units on November 16. Results of other liver function studies remained normal. Electrocardiograms taken on July 16, October 23 and November 4 are shown in Figures 1, 2 and 3, respectively. On July 16 the T wave was inverted in lead II and slightly inverted in lead aVF. On October 23 the T wave in lead II was inverted and more deeply inverted in aVF. On November 4 the electrocardiogram resembled the tracing of July 16. A biopsy specimen of the liver showed mild fatty metamorphosis and focal aggregates of chronic inflammatory cells within the liver lobules. (Fig. 4.) An array of psychological tests showed his I.Q. to be in the dull normal range. An electroencephalogram was normal. It was the opinion of the neurologist that the patient had residuals of heatstroke with diffuse damage to the central nervous system. No evidence of renal abnormality could be found. Other than the electrocardiographic changes there was no evidence of cardiac damage. Fluoroscopy of his heart revealed no abnormalities. Sweating ability was also normal.

Gradually, his limp subsided until it was barely noticeable. He complained of headache, intermittent aching in his arms and legs, and a persistent aching in his right ankle. Persistent frontal headache, which occasionally had a burning quality, was the most prominent symptom. Aspirin, APC, codeine, morphine (given intravenously during liver biopsy), reserpine, Cafergot,® Dilantin® and Octin,® separately and in combination, had no effect. Chlorpromazine (up to 600 mg. per day) was given over an eleven-day period with no change in his symptoms and with no rise in his serum alkaline phosphatase. Seconal,® 50 mg. given three times during the day and 100 mg. at bedtime, gave partial relief for two to three-hour periods. No abnormality could be found to explain the pain in his right ankle. The musculature and skeleton were intact. There were no peripheral vascular or neurological lesions. As his gait improved, the aching gradually decreased in frequency, duration and severity. He still had headaches at the time he was discharged from the hospital but these were slightly less intense. The patient adjusted well, however, to the idea that his headaches might persist for a more or less indefinite period. When he was re-examined in February 1959 there were no changes in his symptoms, physical status, laboratory tests, or liver biopsy specimen.

COMMENTS

Heatstroke Due to Natural Hyperpyrexia. In the many reported series of cases [*3–10*] and in single case reports [*11–17*] of heatstroke, totaling 312 patients, no mention has been made of jaundice or liver disease. Table III tabulates cases of heatstroke in which jaundice and/or liver disease has been recorded. Malamud, Haymaker and Custer [*1*], in an analysis of 125 fatal cases of heatstroke made at the Army Institute of Pathology in 1946, mention that the liver is generally congested and perisinusoidal edema is present. Patients who survived less than thirty hours showed no evidence of hepatic damage. In twelve cases moderate to severe centrolobular necrosis was seen, the earliest being thirty-one hours after onset of the heatstroke. They mention that some livers were relatively ischemic rather than congested. Four patients had had jaundice prior to death. Two other patients had clinically inapparent jaundice but had elevated icterus indices. Mendeloff and Smith [*18*] edited a clinicopathological conference in which a twenty-nine year old long-distance runner collapsed after a six-mile foot race on a very humid day, became unconscious, had a temperature of 103.1°F., and was alternately maniacal. He became icteric on the fifth hospital day, azotemia developed, and he died on the tenth hospital day with a temperature of 105.8°F. There was extensive centrolobular necrosis of his liver. The diagnosis of viral hepatitis with fulminating exacerbation due to heat exhaustion was made because he had been exposed to people with viral hepatitis in the course of his work as a social worker, and because of the histologic picture. He also had lower nephron nephrosis which occurs in heatstroke [*1*]. Nevertheless, this picture is certainly compatible with the liver disease of hyperpyrexia described by Malamud, Haymaker and Custer [*1*] and by Gore and Isaacson [*10*].

Austin and Berry [*2*] analyzed 100 cases of heatstroke and did not mention jaundice. Fifteen van den Bergh determinations were made and in 33 per cent of these results were greater than 1.0 mg. per 100 ml. of blood. Bromsulphalein retention was measured four times and was greater than 20 per cent. The serum alkaline phosphatase was determined eleven times and was always less than 4 King-Armstrong units. Prothrombin times were determined eleven times and were always less than 50 per cent. The pathological findings in the seventeen fatal cases of the total of 100 cases are not given.

Logue and Hanson [*20*] described a twenty-seven year old white man with heatstroke with a temperature of 109°F. On the third day he had jaundice with an icterus index of 57, 3-plus cephalin flocculation test, serum albumin of 4.0 gm. per 100 ml. of blood and a serum globulin of 2.4 gm. per 100 ml. of blood. Ascites developed and the liver and spleen were palpable. His icterus index gradually returned to normal over a one-month period and he eventually recovered completely. Freedman and Schenthal [*21*] describe a ship's engineer in whom, while in a boiler room, heatstroke developed with a temperature of 105.5°F. Coma and diarrhea occurred, and on the second day his liver became enlarged. On the third day he had albuminuria, hematuria, icteric blood serum, and a prothrombin time of 20 per cent. He remained comatose for four days. His liver enlarged to 3 fingerbreadths and he was clinically jaundiced. He recovered completely from his hepatic dysfunction in two months. Permanent neurologic damage remained, as will be discussed. Wright, Reppert and Cuttino [*22*] analyzed twelve cases of heatstroke in which there were six fatalities. Eight of the patients

TABLE III

SUMMARY OF CASES OF NATURAL OR ARTIFICIAL HYPERPYREXIA WITH EVIDENCE OF JAUNDICE AND/OR LIVER DAMAGE

Author	No. of Cases	Overt Jaundice	Hyperbilirubinemia Only	Liver Damage Found by Histologic Study	Fatal Cases	Jaundice and Death
Natural Hyperpyrexia						
Malamud et al. [1]	125	4	2	12	125	4
Austin, Berry [2]	100	...	5	..	17	..
Mendeloff et al. [18]	1	1	..	1	1	1
Logue, Hanson [20]	1	1	..	..	...	..
Freedman et al. [21]	2	1	..	..	...	..
Wright et al. [22]	12	1	5	2	6	1
Saffert [23]	1	1	..	..	1	1
Wilbur, Stevens [30]	1	1	..	1	1	1
Freedman, Rourke [38]	1	...	1	..	...	..
Herman, Sullivan	1	1	..	1	...	..
Total	245	11	13	17	151	8
Artificial Hyperpyrexia						
Gore, Isaacson [19]	16*	5	..	7	16	5
Hartman, Major [27]	2	...	..	2	2	..
Chunn et al. [28]	1	1	..	1	1	1
Warren et al. [29]	283	15	..	..	1	..
Wilbur, Stevens [30]	2	...	..	2	2	..
Wilson, Doan [31]	4	1	2	..	...	..
Brown [32]	1,100	2	..	..	11	..
MacDonald [33]	250	48	..	..	...	..
Wallace, Bushby [34]	254	37	16	..	2	..
King [35]	418	41	30	..	1	..
Watts [36]	1	...	..	1	1	..
Bragdon [37]	404	24	..	1	1	1
Freedman, Rourke [38]	1	1	..	..	...	..
Hartman [39]	1	...	..	1	1	..
Kopp, Solomon [41]	8	1	..	1	2	..
O'Brien et al. [42]	163	...	8	..	...	..
Total	2,908	176	56	16	41	7
Grand Total	3,153	187	69	33	192	15

* Case 17 is the fatal case described by BRAGDON, J. H. [37].

had decreased prothrombin times, three had abnormal hippuric acid tests, and one had mild icterus. In two patients hepatic necrosis was found. One of these patients, an eighteen year old white man, had moderately severe fatty degeneration of the liver. The other, a twenty-five year old white man, had central necrosis of the liver with 60 per cent of the parenchyma destroyed. Saffert [23] described a fifty-six year old white man with heatstroke who remained hyperpyrexic for eight and a half days. His course was complicated by pulmonary edema. He was jaundiced during the last three days before death. No autopsy was performed.

Heatstroke Due to Artificial Hyperpyrexia. In contrast to the paucity of data concerning jaundice in heatstroke, there is much more elaborate material available on jaundice occur-

ring in artificial hyperpyrexia. A number of deaths are described in which no jaundice occurred, and these are indistinguishable from deaths seen in naturally occurring heatstroke [*24–36*]. Artificial hyperpyrexia was employed primarily for the treatment of gonorrhea, particularly the sulfa-resistant forms and also for syphilis of the central nervous system and other diseases. Reported instances of jaundice and/or liver damage occurring in artificial hyperpyrexia are tabulated in Table III. As is shown, various authors [*19,27–41*] have reported at least 176 patients with clinical jaundice, and in nine more liver disease was demonstrated pathologically.

In 1935 Hartman and Major [*27*] reported two deaths from hyperpyrexia. The first patient was a twenty year old pregnant white woman with acute salpingo-oophoritis. She was given three five-hour artificial fever treatments at temperatures ranging from 103° to 106.6°F. Seven days after the last treatment she had a generalized tremor; three days later she had a convulsion, went into coma, and died. Although jaundice did not occur, autopsy showed acute parenchymatous degeneration of the liver and kidneys.

The second patient was a forty year old man with syphilis of the central nervous system who had eleven artificial fever treatments. Following the last treatment he could not be roused and he died twenty hours later. At autopsy, in addition to syphilitic aortitis and right coronary artery ostium stenosis, there was a glioma of the brain with cyst formation, "hemorrhagic encephalitis" of the base of the brain, and acute parenchymatous degeneration of the liver and kidneys.

Hartman [*21*] described a third case—that of a thirty-one year old white man who was treated with artificial hyperpyrexia for iridocyclitis. He died twenty hours after the fourteenth treatment and, in addition to cerebral edema and cerebellar necrosis, the kidneys and liver showed parenchymatous degeneration.

Chunn and Kirkpatrick [*28*] described a thirty year old white man who received heat therapy for gonorrhea for five hours at 107°F. Persistent vomiting occurred, there was a progressively deepening jaundice, and the patient died about 100 hours after hyperpyrexia. He had digested blood in his stomach and small bowel. His liver weighed 1,320 gm., showed central necrosis, and in some blocks of tissue there was almost complete destruction with only an occasionally recognizable bile duct. At the periphery of the lobules a few chains or clusters of deeply stained liver cells were seen.

Warren, Scott and Carpenter [*29*] treated 283 patients an average length of 14.8 hours at 106.7°F. Jaundice and severe nausea occurred in fifteen patients, the icterus index rising as high as 18 by the third day after treatment. The jaundice subsided in three to five days. One death is described, but this patient did not have jaundice.

Wilbur and Stevens [*30*] reported the case of a twenty-three year old white man with gonorrhea who was treated with hyperpyrexia. Two and a half hours after the start of therapy, when his temperature was 106.7°F., convulsions and stupor occurred. Hematuria also occurred and his electrocardiogram, " . . . showed a slight abnormality suggesting myocardial damage." On the third day after treatment he had anesthesia and paralysis of both lower extremities, bilateral papilledema, bilateral subconjunctival hemorrhages, and hemorrhagic cerebrospinal fluid. He died on the third day. Autopsy showed a 2,350 gm. liver with early diffuse necrosis. The kidneys were swollen and had petechial hemorrhages in the pelves. The renal tubular epithelium showed early necrotic changes. There were subendocardial and myocardial petechial hemorrhages. Some of the muscle fibers of the heart and diaphragm were hyalinized. A second patient, a twenty-eight year old white man, was admitted with a urethral discharge and hematuria, and gram-negative intracellular diplococci were found on smear. According to his past history, every hot summer he had three or four heatstrokes which made him unconscious for about three hours. During hot weather he frequently had tinnitus, scotomas and vertigo. He was given artificial hyperthermia and his temperature maintained at 106.7°F. His temperature then rose to 107.6°F., he went into shock, had convulsions, and became stuporous. He died twenty-three hours later. Autopsy showed subendocardial and myocardial petechial hemorrhages. The alveoli of the lungs were filled with red blood cells. The liver weighed 2,300 gm. and showed early diffuse necrotic changes. The kidneys showed early necrosis of the epithelial cells. Astrocytes of the brain were swollen. A third case of heatstroke occurred in a thirty-three year old white man. His temperature was 110°F. He had albuminuria. He lived for three days and his temperature remained above 103°F. He re-

mained stuporous and a few hours before death became jaundiced. Autopsy showed early necrotic changes in the liver and epithelial cells of the kidney tubules.

Wilson and Doan [*31*] showed that in four young adults given fever therapy for gonorrhea results of the bromsulphalein test were above normal and one of the patients became clinically jaundiced with an icterus index of 70, although another patient had only an elevated icterus index. There was some decrease in the prothrombin time of each of the four patients, but in only one patient did the prothrombin time become less than 35 per cent. This patient had ten hours of fever therapy at 105°F. His prothrombin time became 11 per cent and hematemesis occurred. These authors noted that the platelet count decreased to one-sixth to one-fourth of normal and remained decreased for forty-eight to seventy-two hours. In one patient epistaxis occurred during the period of thrombocytopenia.

Brown et al. [*32*] gave 1,100 hyperthermic treatments with temperatures of 106.7°F. There were eleven deaths among this group of patients. He lists two non-fatal cases in which jaundice occurred. MacDonald [*33*] treated 250 patients who had sulfa-resistant gonorrhea and encountered forty-eight instances of jaundice. Five patients had marked jaundice, six had moderate jaundice, and thirty-seven had mild or doubtful jaundice. All the doubtful patients had an elevated icterus index, usually about 15 or 20 units. In a few cases the liver was considered to be enlarged but never greatly so. Tenderness in the area of the liver was encountered only a few times.

The usual course of jaundice in these patients is described as follows: Within twenty-four hours of the fever therapy, nausea and vomiting begin. The urine becomes dark, and jaundice appears in forty-eight hours; it usually disappears within a week. Wallace and Bushby [*34*] treated 254 patients with sulfa-resistant gonorrhea at a temperature of 106.6°F. for eight hours. Jaundice appeared in thirty-seven of these patients. Vomiting was particularly evident in cases of clinical jaundice. Icterus developed in one group of their patients twenty-four to ninety-six hours after therapy and persisted for a few days. Another group of seven patients had an elevated serum bilirubin and three were clinically icteric. Drowsiness, stupor and disorientation were noted in this group. In a third group of eleven patients icterus occurred in two, and the serum bilirubin was seen to rise as early as four hours after termination of the treatment. In a fourth group of forty-two patients there were three cases of jaundice, and in a final group of nineteen patients eighteen had elevation of serum bilirubin and three were clinically jaundiced. Eight patients were given benzoic acid after heat therapy and all showed decreased excretion of hippuric acid which was interpreted as showing impairment of liver function.

King et al. [*35*] gave 758 treatments to 418 patients. Jaundice occurred in forty-one patients but was usually mild and transient. Of forty patients in whom the serum bilirubin was measured before and for three days after treatment, thirty had a rise in serum bilirubin above 0.6 van den Bergh units, being maximal on the first day after treatment. Intravenous hippuric acid tests were performed in forty patients before and on the third day after treatment. Twelve of these patients gave an abnormal response. There was one fatality in this group (the authors had another death in a group of patients not included in this study).

Watts and Hartman [*36*] described a patient with multiple sclerosis who, after four or five hyperpyrexia treatments, had vascular collapse and lived only thirty-six hours. The autopsy findings were: focal necrosis in the liver, brain edema, cerebral and pulmonary congestion, and thrombosis of the lateral sinuses. Bragdon [*37*] in 1947 gave 404 patients with sulfa-resistant gonorrhea 524 treatments with fever at 106° to 107°F. for seven hours. Jaundice developed in twenty-four of their patients, and one patient died. This latter case is described in detail. After four hours of therapy this patient's rectal temperature was 108°F. He became unconscious, ceased to sweat, had bilateral tonic movements of his extremities, and was in shock. When ice packs were applied he became alert, his temperature returned to normal, and neurological signs disappeared. Twenty-four hours later, however, nausea and vomiting appeared and forty-eight hours after onset he was jaundiced, and his liver was palpable and tender. Over the next seven days his liver shrank and his jaundice deepened. Gross hematuria, oliguria, uremia and anasarca developed, but in the second week the hematuria ceased and diuresis occurred. His serum non-protein nitrogen decreased and he became rational. Vomiting persisted, however, and on the fourteenth day

he had a massive, fatal bout of hematemesis. At autopsy, in addition to jaundice, anasarca was present and 2,200 ml. of ascitic fluid was found in the abdomen. Acute ulceration was evident over the lower esophagus and was thought to be due to the prolonged nausea and vomiting and to be the probable site of the hemorrhage. His liver weighed 1,800 gm. and the parenchyma was yellow and friable. In the liver, relatively acellular areas were found alternating with areas of regenerating liver cells. The acellular areas consisted of collapsed, edematous stroma with occasional liver cells. Over 70 per cent of the liver had areas of regeneration. In the kidney, the distal tubules showed evidence of necrosis and regeneration.

This case was also included as Case 17 in the series of seventeen deaths due to artificial hyperpyrexia analyzed by Gore and Isaacson in 1949 [*19*]. Jaundice occurred in five other cases in this series and renal failure in two. Seven patients had central necrosis of the liver while fatty vacuoles were found in thirteen. Neuronal damage was found in fourteen of the fifteen patients in which this was looked for. There was fatty degeneration of the heart in four cases, granular and hyaline degeneration in seven, and edema in three others. Nine of the sixteen patients showed proximal tubular degeneration. Four more had lower nephron nephrosis. Icterus occurred uniformly in all patients who survived longer than forty-nine hours. Congestion and hemorrhage were striking features in these cases.

The composite pathological picture of the liver was described as follows: The earliest change in the liver was "cloudy swelling." At eight hours after hyperthermia, tiny vacuoles were seen in the cytoplasm—so-called watery vacuolization. By ten hours, small fatty droplets appeared in the cytoplasm, coalesced, and became larger. Centrolobular necrosis appeared, minimal at sixteen hours but by sixty hours 60 per cent of the central part of the lobule had become necrotic and hyperemic. At seven days, phagocytosis by macrophages was seen and the relatively undamaged cells at the periphery of the lobule underwent active regeneration. Proliferation of biliary channels occurred simultaneously. "Morphologically, such a liver may resemble those found in fatal cases of epidemic hepatitis, but the sparsity of inflammatory cellular reaction and the lack of liver atrophy are helpful differential features." No appreciable fibrosis was present [*19*].

Freedman and Rourke [*38*] treated a twenty-two year old man, who had sulfa-resistant gonorrhea, with artificial hyperpyrexia by administering typhoid vaccine intravenously. After the third injection his temperature rose to 110°F. He had a convulsion but thereafter his condition gradually improved. On the third hospital day icterus developed, with an icterus index of 19 and a prothrombin time of 50 per cent. Only permanent neurological damage remained, as will be discussed. These authors also describe a sixty-three year old man with high fever and delirium due to pneumonia. His highest temperature was 106°F. His skin was hot and dry, he had albuminuria, was semicomatose, and had a total serum bilirubin of 1.7 mg. per 100 ml. of blood, bromsulphalein retention of 9 per cent, and his liver was enlarged 3 fingerbreadths. He recovered but sustained permanent neurological residuals as mentioned herein.

Levy [*40*] described 2,929 hyperthermic treatments given to 1,694 patients with temperatures of 106°F. for eight hours. Only two deaths occurred. One of the complications listed was " . . . subclinical jaundice sometimes progressing to the clinical level." No other mention of jaundice was made. Kopp and Solomon [*41*] reported eight cases of shock during artificial fever therapy, with two deaths. In one autopsy case cloudy swelling of the liver was found. In one patient, who survived shock and had jaundice for eight days beginning on the third postfever day, the icterus index rose to a maximum of 85.0. O'Brien et al. [*42*] treated 163 patients with drugs and artificial fever, and eight patients had elevated icterus indices.

The jaundice in our patient occurred on the third hospital day after consciousness had returned. Liver damage intimately associated with heatstroke is the most likely explanation. One cannot rule out entirely the possibility that a latent infectious hepatitis was precipitated by the heatstroke or that it occurred coincidentally. It is well known that viral hepatitis can occur in the form of intrahepatic obstruction [*43*]. Yet this does not seem to be a likely explanation in view of the experiences of others, as previously mentioned. Another possibility could be that the jaundice was due to intrahepatic obstruction following administration of chlorpromazine. Sussman and Sumner [*44*] described a patient with jaundice after a single 50 mg. dose of chlorpromazine. When their patient was given

a disguised test dose of chlorpromazine seven months after the initial dose she became jaundiced in three days. Giambescia et al. [*45*] had eight patients with chlorpromazine jaundice. One of these patients had chlorpromazine hepatitis in November 1953 and again in April 1954. He had a recurrence in February 1955 following reinstitution of the chlorpromazine therapy. Hollister [*46*] challenged eleven patients who had had jaundice due to chlorpromazine therapy, ten days to seventeen months after the initial episode of jaundice. Clinical jaundice occurred in six. Two patients had fever and gastrointestinal symptoms with increased bromsulphalein retention and elevated alkaline phosphatase but without jaundice. One patient had fever and vomiting but no jaundice. Liver function tests were not obtained in this latter patient. Two patients showed no reaction. One patient was first given chlorpromazine a little less than five months after the onset of his jaundice (about three and a half months after disappearance of his jaundice). He received 300 mg. per day for three days, 400 mg. per day for five days, and 600 mg. per day for three days. He remained afebrile, had no gastrointestinal symptoms, showed no icterus, and his alkaline phosphatase remained unchanged. Although this does not absolutely eliminate chlorpromazine as an etiological factor, it makes it rather unlikely.

Malamud et al. [*1*] demonstrated that hemorrhage is an important pathological event in heatstroke, with cerebral hemorrhage, petechiae and purpura of the skin, subendocardial and subepicardial hemorrhage, hemorrhage into cardiac muscle, pulmonary hemorrhage, and hematuria. Our patient demonstrated diffuse and profound neurological changes and had petechiae and purpura of his skin, and subconjunctival hemorrhage of the eye, but he did not show evidence of pulmonary hemorrhage or hematuria. His abnormal electrocardiogram may indicate residuals of intracardiac hemorrhage. Resolution of such hemorrhagic deposits might explain some of his jaundice, as might the presence of a hemolytic process. Halden et al. [*5*] studied fifteen patients with heatstroke, in whom anemia developed in one week but who had no obvious blood loss. Ten of these patients had an increased rate of red blood cell destruction as measured by radiochromium-tagged cells. Four-day stool urobilinogen measurements averaged 242 mg. per day in six patients. The authors concluded that "the anemia of heatstroke is partly due to a hemolytic state and partly due to injury to the bone marrow." No mention was made of jaundice in any of their patients. A hemoglobin of 12.0 gm. per cent did develop in our patient as compared to prior consistent values of 15.7 gm. per cent. At a time when his total serum bilirubin was 14.2 mg. per 100 ml. of blood, his fecal urobilinogen was 166 Ehrlich units per 100 gm. and a reticulocyte count was 0.7 per cent. In favor of parenchymal liver damage is a serum transaminase of 131 units which fell gradually to 42.5 units and finally to 27.5 units. Later, bromsulphalein retention of 10 per cent was recorded with a serum transaminase four days later of 50 units. Twenty-five days later the serum transaminase was 24 units. Early in December his bromsulphalein retention was 4 per cent. A biopsy specimen of the liver showed fatty metamorphosis. Administration of chlorpromazine failed to produce any sign or symptom of hepatitis. The clinical course of the icterus with regard to time of onset, persistence, liver function tests and recovery was indistinguishable from that seen in other cases of hyperpyrexia either naturally occurring or artificially induced. Therefore, it is our thesis that this patient represents an instance of jaundice due to liver damage secondary to heatstroke.

No clinical evidence of cardiac disease appeared in this patient. Yet electrocardiograms obtained on July 16, October 23 and November 4 showed abnormalities. The electrocardiogram of July 16 showed an inverted T wave in leads III and aVF while in that of October 23 T wave inversions were more pronounced in leads II, III, and aVF while the electrocardiogram of November 4 showed less T wave inversion in leads III and aVF. The T waves in the precordial leads became taller in this last electrocardiogram. Malamud, Haymaker and Custer [*1*] recorded an electrocardiogram showing a changing pacemaker and inverted to upright T waves in leads II and III. Pathologically, considerable damage was found in the hearts studied by Malamud et al. [*1*], consisting mainly of hemorrhage in the cardiac muscle in twenty-four cases and large subendocardial hemorrhages in ten. Degenerative changes of the muscle fibers were found in thirteen of twenty-seven cases in which the illness lasted more than twenty-four hours. Fragmentation and rupture of muscle fibers as well as fatty changes were found. Logue and

Hanson [*20*] described a patient with heatstroke and jaundice who displayed electrocardiographic abnormalities. On the eleventh day, sinus bradycardia and T wave inversion in leads I, II, III and IV (CF-4 probably) was seen. On the twenty-first day the T waves in lead IV were deeper but were upright in the other leads. Two months later the T wave in lead I was slightly inverted, in lead II it was diphasic, and and in leads III and IV it was inverted. Three months later the electrocardiogram was normal. Since our patient had evidence of bleeding initially (purpuric spots on his extremities and subconjunctival hemorrhage), it is possible that residuals of some intracardiac hemorrhage account for the electrocardiographic changes. The serum transaminase of 131 units was too high to be due to cardiac changes and in all probability was due to the hepatic damage.

The profound neurological disturbance occurring initially is in keeping with the pathological findings of Malamud et al. [*1*] who found severe edema of the brain with degeneration of cortical neurons and increase in the glia in the cortex. Similar damage was present in the corpus striatum and thalamus, and there was diffuse damage to the caudate nucleus and putamen. The cerebellum showed severe changes with edema of the Purkinje layer, and damage of and a decrease in the number of Purkinje cells. Patients who survived more than twenty-four hours showed almost complete degeneration of the Purkinje layer with severe gliosis. Gore and Isaacson [*19*] found similar changes in the central nervous system. The brain showed perivascular edema. Parenchymal damage was most striking in the cerebellum, with focal neuronal degeneration in the Purkinje cell layer when death occurred in less than twenty-four hours. With survival beyond twenty-four hours there was edema and glial proliferation. Cortical neuronal damage was seen, but glial reaction was not. In Case 10 definite areas of ischemic necrosis were most prominent in the deeper layers of the cerebral cortex and in the periventricular part of the thalamus.

Several cases of permanent cerebellar dysfunction following heatstroke have now been reported. Freedman and Rourke [*38*] had two cases of heatstroke. One occurred during artificial fever therapy for sulfa-resistant gonorrhea and another less clear-cut case was due to fever in pneumonia. Both patients had permanent dysarthria and ataxia after recovery from the acute phase of their illnesses. Similar permanent cerebellar disease following heatstroke has been reported by Freedman and Schenthal [*21*] (two cases), Cruickshank [*16*] (one case), Silverman and Wilson [*17*] (one case), McAlpine [*47*] (three cases), Walter [*10*] (three cases), Freeman and Dumoff [*48*], and Stewart [*49*]. Weisenberg [*50*] reported one case and cited two cases reported by Nonne and other cases reported by Goebel, Revenstorf and Hiller. The patient of Logue and Hanson [*20*] exhibited nervousness and slight tremor of the hands during convalescence and his electroencephalogram showed abnormalities which gradually disappeared. It is interesting that this patient's nervousness and slight tremor were attributed to psychoneurosis and no mention was made of the possibility of cerebellar involvement. The selective sensitivity of the Purkinje cells of the cerebellum to hyperpyrexia has been well demonstrated histologically by Malamud et al. [*1*] and by Gore and Isaacson [*19*].

These changes explain the neurological picture in the present patient. The initial changing neurological picture in this patient can be well equated to the changes in degree and extent of edema of the cortex and subcortical structures. The later disturbances in gait and motor control again are consistent with the expected residual gliosis and cellular damage that would be expected to have taken place and may well have been due to cerebellar damage. Finally, the persistent headache and aching of the extremities can be explained on the assumption of predominant residual damage of the thalamus. But this patient did not demonstrate severe burning pain increased by movement or repetitive stimulation of the painful areas except that he found it necessary to limp on his right leg because of the pain. There were no vasomotor or trophic disturbances. According to his parents, his personality had changed from that of a pleasant, placid individual to a rather uncommunicative, continuously complaining individual. His pain was not relieved by the administration of any of the usual analgesics. Thus there are similarities to the thalamic syndrome, but the picture is not identical with it.

Little mention has been made in the literature of the residual personality changes which take place in heatstroke. Gauss and Meyer [*4*] noted that in some patients on recovery from coma there was a marked disturbance of speech and a few would not or could not speak. This

may have been a manifestation of the cerebellar dysfunction previously mentioned. Some had an inability to hold their attention to the doctor's questioning. Some had loss of orientation for time and place and a few had hallucinations and delusions. No follow-up has been made of survivors of heatstroke in any of the cases cited, however, Chapman and Bean [*14*] had a patient with heatstroke following bronchoscopy. His temperature was 109°F.; he went into a coma, had signs of decerebrate rigidity, and was restored in one week to his previous state of health but he complained of "fantastic dreams." Wakefield and Hall [*51*] in their excellent historical review of heatstroke briefly cite the case of a forty-seven year old white man who had heatstroke fifteen years prior to being seen by them. Since that first heatstroke he had had frequent headaches, visual disturbances, and an inability to maintain his balance with his eyes closed. When seen he had absent knee jerks, dilated pupils which did not respond to light, and a positive Romberg sign, but no evidence of syphilis. Margolis and Gianascol [*52*] described a patient with the thalamic syndrome of burning pain who improved and maintained improvement for sixteen months on a regimen of 400 mg. of chlorpromazine per day. Our patient was treated with chlorpromazine for eleven days with increasing doses up to 600 mg. per day without any effect. His only relief has been obtained with the ingestion of Seconal. It is our contention that this patient's disease is explicable entirely on the basis of acute heatstroke with residual damage.

SUMMARY

A patient who suffered heatstroke and jaundice due to liver disease is described. Upon recovery from the acute phase of his illness the patient demonstrated these residuals of heatstroke: fatty metamorphosis of the liver, persistent headache, and aching of the extremities relieved only with the ingestion of Seconal. He had an abnormal electrocardiogram.

Of 3,152 cases of hyperpyrexia recorded in the literature, jaundice was found in 186. There were 191 deaths, and jaundice was present in fourteen of these.

Of 244 cases of heatstroke, there was overt jaundice in ten, hyperbilirubinemia only in thirteen, and of 150 fatalities jaundice was found in only seven cases. Liver damage was found by histological study in seventeen cases.

The liver damage of hyperpyrexia is an occasional and serious complication of heatstroke.

ADDENDUM

Since this manuscript was submitted Schillhammer and Massonneau have published a report which includes a description of the early phases of this patient's illness [*53*].

Baxter and Teschan [*54*] have reported three fatal cases of heat stroke; centrolobular necrosis of the liver was present at postmortem examination in each.

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Dissolution of Renal Uric Acid Stones by Oral Alkalinization and Large Fluid Intake in a Patient Suffering from Gout*

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THE ideal management of renal calculi is their dissolution *in vivo*. Spontaneous dissolution of renal calculi has been observed but apparently is rare [*1*]. More numerous are the cases in which dissolution of kidney stones was achieved by treatment in one form or the other, such as large fluid intake combined with acid ash diet for calcium phosphate and calcium carbonate stones [*2,3*] or with alkalinization for cystine stones [*4*]. Dissolution of uric acid stones in the kidney by medical treatment apparently has been only rarely achieved. Prien [*5*], Colby [*6*], Greene [*7*] and some physicians who replied to Burkland and Rosenberg's questionnaire on urolithiasis [*8*] mention dissolution of uric acid stones by medical treatment, but no documentation was given. It is not mentioned in several standard textbooks of urology [*9–11*] or modern monographs on renal lithiasis [*12*].

In the present communication we report on a patient suffering from gout, in whom uric acid stones were dissolved by medical treatment within one year or possibly even within five months. This result is the more striking as it was achieved while the patient was being treated with a uricosuric drug.

CASE REPORT

S. S., a fifty-four year old Russian born engineer, living in Israel since 1925, and the father of two healthy children, was admitted to the medical department on January 8, 1957, with gouty arthritis and bilateral renal colic.

The past history revealed scarlet fever at the age of six and relapsing fever and malaria in the years 1925 to 1935. In 1948 he had his first attack of gout in the metatarsal-phalangeal and interphalangeal joints of both great toes. Since then he had had numerous attacks, involving the distal and proximal joints of the lower and upper extremities and the right sternoclavicular joint. Tophi at the wrists and in the left ear cartilage were first noted in 1951. Since 1952 he had chronic pains in many joints, with severe limitation of movement. He was treated sporadically with salicylates, colchicine and atophan, and during the years 1954 to 1955 he received 1.5 gm. of probenecid daily. In the course of that year his attacks decreased in frequency and severity; however treatment with probenecid was discontinued because of difficulty in obtaining the drug.

In 1949 he began to suffer from recurrent left renal colic, with hematuria. Since 1951 he had recurrent right renal colic. In 1953 he passed a small stone, which was not examined.

In 1955 he was admitted for urologic examination to another hospital, where the following findings were recorded. The urine contained a trace of protein, numerous red cells and a few granular casts; the pH was 5.0, the maximum specific gravity 1.010. The blood urea was 27 mg. per cent; serum uric acid, 8.5 mg. per cent; and serum calcium, serum inorganic phosphorus and alkaline phosphatase were normal. The urea clearance was 65 per cent of normal. Intravenous urogram revealed good filling of the right renal pelvis and calyces with a normal renal outline, and poor excretion by the left kidney, which was visible only on tomography. Cystoscopic examination combined with intravenous injection of methylene blue showed normal excretion in the right kidney and the appearance of a small amount of uncolored urine in the left kidney. Retrograde pyelography of both kidneys was normal. After discharge from the hospital the patient continued to suffer from severe colic, mainly left sided, with recurrent hematuria and severe attacks of arthritis with progressive disability.

On admission to our hospital in January 1957, the patient was in fair nutritional condition. Tophi were noted in the cartilages of both ears and on the left

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FIG. 1. A, excretory urogram on January 10, 1957, showing a filling defect occupying most of the right renal pelvis and another filling defect in the caudal part of the pelvis due to non-opaque stones. The calyces are dilated. B, right retrograde pyelogram on January 22, 1957. A large filling defect is seen in the dilated pelvis, surrounded by a narrow opaque zone. A second smaller filling defect is visible in the caudal calyx. The ureter is dilated.

wrist. There was severe limitation of movement in nearly all the joints of the extremities. The metatarsalphalangeal joints of both toes were red, swollen and tender. There was marked tenderness in the left loin. Further physical and neurological examination was essentially within normal limits. The systolic blood pressure was 130 mm. Hg; the diastolic pressure, 90 mm. Hg. The urine contained a trace of protein, numerous red cells, a few white cells, a few granular casts and many uric acid crystals; the pH was 5.4, the maximum concentration 1.019; the urinary uric acid excretion, while the patient was given a normal hospital diet, was 460 to 600 mg. per twenty-four hours. Paper chromatography for amino acids was normal. The hemoglobin was 12 gm. per cent. Blood chemical studies included normal values for urea, glucose, calcium, inorganic phosphorus, chloride, sodium, potassium, albumin, globulin and alkaline phosphatase. The serum uric acid was 7.6 mg. per cent. The urea clearance was 55 per cent of normal.

Roentgenological examination showed a few well defined bone erosions near the surfaces of the metatarsophalangeal and the interphalangeal joints of both toes and the right elbow joint, with degenerative arthritic changes.

Excretory urogram was performed on January 10, 1957. (Fig. 1A.) The plain film revealed no abnormalities. Five minutes after injection of 20 cc. of urognost (diodone, 35 per cent),* both the pelves and calyces of both kidneys were well outlined with good function.

* Assia Chemical Lab. Ltd., Tel Aviv, Israel.

The left kidney pelvis and calyces were normal. The right kidney showed marked dilatation of the pelvis, as well as of the calyces; the fornices were wide and plump, and could hardly be differentiated from the calyces. In contradistinction to the good contrast of the calyces, the pelvis was radiotranslucent; a filling defect occupying most of the renal pelvis and an additional discrete filling defect in the caudal part of the pelvis were seen. The ureteropelvic junction did not show the normal funnel shape but had a concave border towards the pelvis.

As non-opaque stones in the right pelvis and lower calyx were suspected, a right retrograde pyelogram was taken on January 22, 1957. (Fig. 1B.) The ureter was dilated, grade 1. The beginning of the ureter lay above the lower border of the pelvis. The pelvis was more radiotranslucent than the calyces and bordered by a narrow opaque zone. In the caudal calyx a well defined translucent spot about the size of a pea was visible, while the clear zone in the caudal part of the pelvis, seen in the intravenous urogram, had disappeared. The roentgenological diagnosis of a large non-opaque stone in the pelvis and a smaller one in the caudal calyx was made. The possibility of the translucency being caused by gas in the intestines could be excluded as the radiotranslucent zones in the kidney pelvis appeared in the intravenous as well as in the retrograde urogram, and in the latter the translucent zone was surrounded by a narrow circular band of contrast medium filling the small space between the surface of the stone and the pelvic wall.

TREATMENT AND COURSE

The large stone in the right kidney pelvis, with marked hydronephrosis, posed an urgent therapeutic problem. From the urologist's point of view surgical removal of the stone was indicated without delay. The internist, on the other hand, considering the stones to be composed of uric acid, thought a trial of *in vivo* dissolution by alkalinization worthwhile, since uric acid stones can be dissolved *in vitro* by alkali (albeit with difficulty), only the soft friable organic matrix remaining [*13*], and since the formation of uric acid stones can be prevented by oral alkalinization therapy [*5,6,14*]. Although the treatment with probenecid, indicated for the severe gout, would increase the urinary uric acid excretion and thus might enhance stone formation, it was decided to postpone surgery and to institute oral alkalinization therapy combined with forcing fluid.

The patient was given probenecid 2 gm. daily, colchicine 1 mg. three times per week, and a low purine diet. A minimal daily fluid intake of 2.5 L. was prescribed and the patient was instructed to drink two glasses of water before retiring and again at two o'clock at night. In addition he received four tablespoonfuls (60 ml.) of an alkalinizing mixture, according to Eisenberg, Connor and Howard [*15*], consisting of citric acid, 40 gm.; sodium citrate, 60 gm.; potassium citrate, 66 gm.; extract aurantii, 6 gm.; syrup simplex ad 600 cc.

The patient's urine was checked periodically for quantity, pH and uric acid crystals. During the period from the end of January until the end of May 1957 his urinary output generally exceeded 1,500 ml., the pH was 7 to 7.3 and no uric acid crystals were found. During this period the urine contained traces of protein, a few granular casts, but no red cells. On two examinations the twenty-four-hour uric acid excretion was 800 and 850 mg. respectively. During the first six weeks of this treatment the patient had several at-

FIG. 2. Excretory urogram on May 26, 1957. The dilatation of the right renal pelvis and calyces has subsided greatly. No stones are visible.

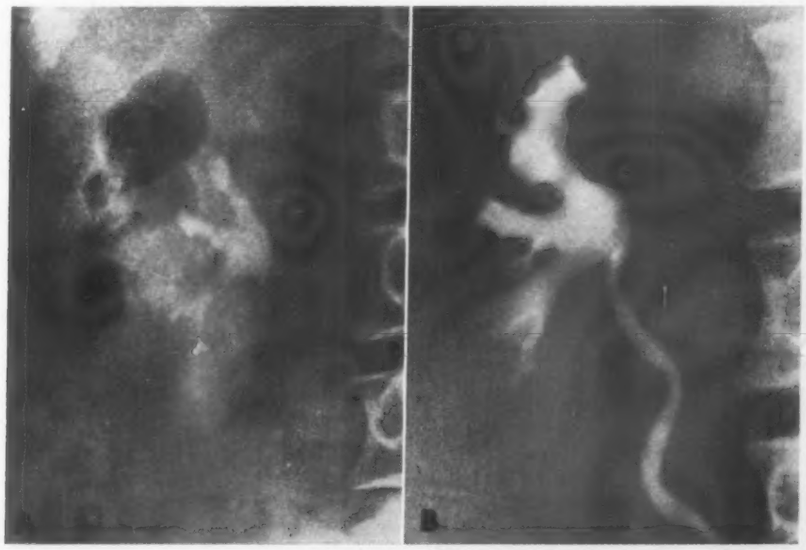

FIG. 3. A, excretory urogram on December 18, 1957. Note the normal width and shape of pelvis. No stones are visible. Ureteropelvic junction and ureter are normal. B, right retrograde pyelogram on January 12, 1958. Pelvis, calyces and ureter are well filled and normal.

tacks of renal colic on the left and a few on the right, but from then on he had no recurrence of colic. After three months he noted marked improvement in his joints, both with regard to the pains and the limitation of movement. The number and severity of the attacks of acute gouty arthritis decreased. The serum uric acid on May 1, 1957, was 6 mg. per cent.

Excretory urogram was repeated on May 26, 1957. (Fig. 2.) The dilatation of the right renal pelvis and calyces had greatly subsided. No translucent spots could be seen. The right ureter at the ureteropelvic junction now had the normal funnel-shape appearance. The tone of the ureter was still diminished. This change in the roentgenogram after three months of treatment does not prove with certainty that the stones had altogether disappeared; however, if still present, they must have considerably diminished in size.

On December 18, 1957, the patient was re-examined. He had no arthritic pains, and joint movements were normal, but the tophi were unchanged. His last attack of acute arthritis occurred in September 1957 and was very slight. Roentgenological examination of the joints did not show improvement. There had been no recurrence of renal colic and no hematuria. The urine still contained some albumin and many granular casts. The blood urea was 57 mg. per cent; a clearance was not performed. The blood uric acid on January 8, 1958, was 4.9 mg. per cent. No stones were passed during the whole period of treatment.

Excretory urogram (Fig. 3A) showed normal width and shape of the right pelvis and calyces. The fornices were pointed and slender, and the ureteropelvic junction was normal. The opacity of the pelvis and calyces was homogenous; no filling defects could be seen. Right retrograde pyelogram on January 12, 1958 (Fig. 3B), showed the pelvis and calyces normal. Additional evidence for the disappearance of the stones is furnished by the ureter having regained its normal tone.

COMMENTS

According to Prien [*16*], Boyce [*17*] and Twinem [*18*], in the United States 4 to 6 per cent of renal calculi are composed of uric acid or contain uric acid in admixture with various salts. In some areas of the United States such as the East, South and Southeast [*8*], and in Southern Asia [*18*] and in Israel [*19*] the incidence of uric acid stones is higher.

Uric acid stones are frequent in primary and secondary gout [*20–22*], lymphomas [*23*] and leukemia [*24*]. In gout, during treatment with phenylbutazolidine [*25*] or probenecid and other uricosuric drugs [*26*], and in leukemia and lymphomas during chemotherapy or radiation therapy [*23,24,27–29*] the formation of uric acid stones or gravel may cause anuria and even death due to blocking of urinary passages. The problem of prevention and cure of uric acid lithiasis is therefore of considerable importance.

The urine contains uric acid and urate in supersaturated solution. With rising pH the proportion of urate and the solubility increase [*30*]. Precipitation of uric acid in the urinary tract occurs when the solubility in the urine is exceeded, due to inadequate urinary volume or low urinary pH. Accordingly, forcing fluid intake and alkalinization of the urine are generally recommended as a precautionary measure during treatment of the aforementioned diseases and for the prevention of recurrent uric acid stones in general.

Although the urine contains both uric acid and urates, Prien has shown [*16*] by crystallographic examination that stones are composed of uric acid and not of urate. Accordingly, Haskins' quantitative data on the solubility of uric acid in human urine [*31*], determined by equilibrating urines of various pH with uric acid crystals, were used in trying to dissolve the stones in our patient. At a pH of 7.0 or above and a minimal daily urine output of 1,500 cc., the urine can contain at least 2,100 mg. of uric acid plus urate [*31*]. Actually the uric acid excretion by our patient during probenecid treatment did not exceed 850 mg. per twenty-four hours. Maintenance of a large margin between uric acid-urate content and solubility seemed desirable, as Haskins' figures based apparently upon measurements in normal urines [*31*], may not hold for pathological urines in which various unknown factors may influence uric acid solubility and uric acid stone formation [*30*].

Prien [*5*] stressed the importance of avoiding even transient periods of oliguria in stone-forming subjects, in order to prevent having the concentration of stone-forming substances in the urine exceed the maximum solubility even temporarily. Special stress was therefore laid upon drinking before retiring and once more during the night, in the management of the present case.

SUMMARY

Dissolution of renal uric acid stones in a patient with gout was achieved by oral alkalinization and large fluid intake.

Acknowledgment: We are indebted to Miss C. Mayer for technical assistance.

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Persistent Lactation Associated with Pituitary Tumor and Hyperadrenal Corticism*

Successfully Treated with Pituitary Radiation

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PERSISTENT lactation is an infrequent finding. It is seen associated with the amenorrhea and hyperinvolution of the uterus following prolonged nursing after childbirth, the so-called Chiari-Frommel syndrome [*1*]. It is also known to occur in acromegaly; and has been observed in some cases of premature menopause, occasionally following oophorectomy, and hysterectomy [*2*].

Recently a syndrome characterized by galactorrhea, amenorrhea, diminished or absent urinary excretion of gonadotropins, and in many cases association with a pituitary tumor, has been described by Argonz and Castillo [*3*] and Forbes et al. [*4*]. In the fifteen cases recorded by Forbes and co-workers all but two patients showed a moderate increase in the urinary excretion of 17-ketosteroids.

We have recently had occasion to study a patient with persistent lactation in whom enlargement of the sella turcica and increased urinary excretion of both 17-ketosteroids and 17-hydroxycorticosteroids were found. Detailed observations on the relationship between lactation and steroid levels, and on the effect of pituitary radiation on these functions forms the basis for this report.

CASE REPORT

V. M., a thirty-eight year old white housewife, was first admitted to the Barnes Hospital in April, 1955. Her physical condition had been normal until shortly after her marriage in 1940 when she had the onset of amenorrhea, gradual weight gain of 40 pounds, and persistent lactation from both breasts. There was no history of mechanical stimulation, and the patient had never been pregnant. There had been no particular increase in body hair. She had been a school teacher prior to her marriage; however, shortly thereafter she began to exhibit symptoms indicative of schizophrenia. These symptoms had increased in severity in the year prior to her admission to the hospital.

On examination the blood pressure was 130/80 mm. Hg; other vital signs were normal. The patient was obese, most of the obesity being limited to the trunk and the face. The skin color was normal with a slight plethora about the face. Slight facial acne and a moderate growth of hair on the upper lip were noted. The hair on the head was coarse. The distribution of body hair was normal. The breasts were large, with palpably enlarged ductal and glandular tissue. A few drops of milky fluid could easily and consistently be expressed from each breast. The cardiorespiratory system was within normal limits. The abdomen was obese. There were no purple striae. The vaginal introitus was small, the clitoris was not enlarged. The uterus was small and there were no adnexal masses. The ovaries appeared to be normal on palpation. The neurological examination was also within normal limits with the exception of a small superior temporal scotoma of the visual fields.

The hemoglobin was 12.3 gm. per 100 ml.; the white cell count was 5,300 per cu. mm., with a normal differential cell count. The serum sodium, potassium and chloride were within normal limits. Urinalysis revealed no abnormalities. Fat globules were seen on microscopic examination of the secretions from the breast. A vaginal smear showed both reduced desquamation and reduced to absent cornification. Lateral radiographic films of the skull demonstrated an enlarged sella turcica compatible with a pituitary tumor. Intravenous pyelograms with presacral air insufflation were normal. Laboratory data relating to endocrine function are recorded in Table 1. The urinary steroids during her first admission are charted in Figure 1.

Because of the pituitary tumor the patient received radiation to the pituitary delivered through three ports (left and right lateral and vertex), a 1,000 r tumor dose to each port being given over a forty-eight-day period. After five days of therapy there was a

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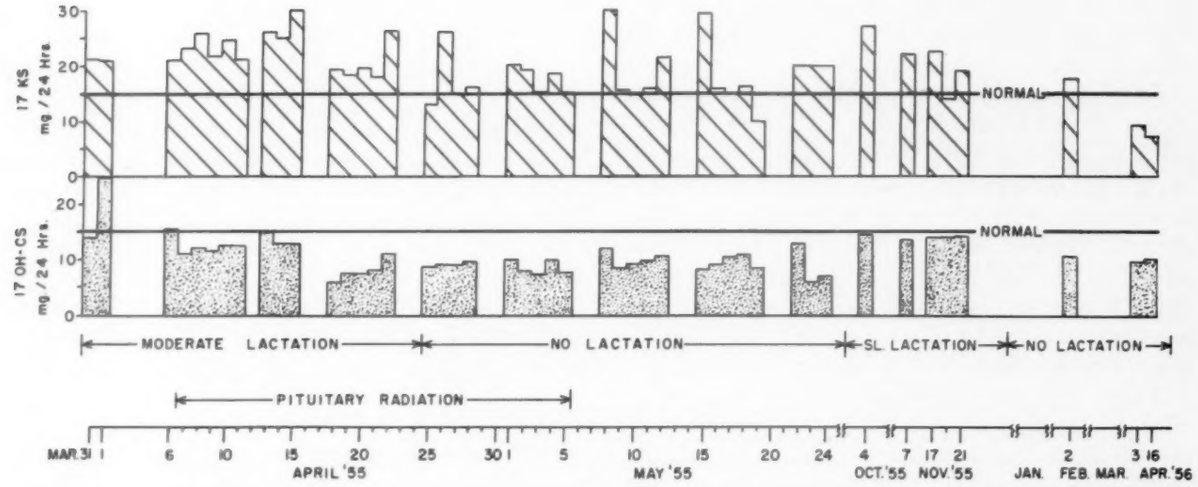

FIG. 1. Effect of pituitary radiation on lactation and urinary excretion of 17-ketosteroids and 17-hydroxycorticosteroids.

definite decrease in lactation, with a corresponding fall in the urinary steroids. (Fig. 1.)

Following completion of radiation therapy the patient was seen about every six weeks. The urinary excretion of steroids remained at high normal levels. In October 1955, six months after radiation therapy, she was having only minimal lactation. The patient was readmitted to the hospital for further study in November 1955. The urinary output of 17-ketosteroids and 17-hydroxycorticosteroids was noted to have increased moderately at this time. (Fig. 1.) The urinary 17-ketosteroids averaged 20.7 mg. per twenty-four hours, the 17-hydroxycorticosteroids averaged 13.7 mg. per twenty-four hours. All other endocrine studies were unchanged, as was a repeat sellar series. The patient still had amenorrhea.

In February and April 1956, the excretion of both 17-ketosteroids and 17-hydroxycorticosteroids was normal. (Fig. 1.) There was no further lactation.

Because of the patient's schizophrenic behavior she was admitted to a state hospital for institutional care. One year later a communication from this hospital stated that she had had no lactation during the past year.

COMMENTS

A clinical classification of the types of abnormal lactation cannot at this time be made on the basis of pathological physiology. Indeed, the same hormonal mechanisms may operate in persistent postpartum lactation (the Chiari-Frommel syndrome) as in the Forbes-Albright syndrome, in which galactorrhea is associated with amenorrhea and usually with a pituitary tumor. In this regard it has become apparent that the Chiari-Frommel syndrome is not, as previously thought, a functional psychiatric disorder but is in fact an organic phenomenon, as demonstrated in recent reports in which a pituitary tumor was found [*5,6*]. Thus except that there is a preceding pregnancy, the Chiari-Frommel syndrome is very similar to the Forbes-Albright syndrome and to our case.

Forbes and co-workers [*4*] placed considerable emphasis on the "low" levels of urinary gonadotropin found in their patients. In the presence of amenorrhea this finding provides evidence only against primary ovarian failure; and is

TABLE I
DIAGNOSTIC STUDIES

Laboratory Procedures	Results
Electrolytes	Normal
Basal metabolic rate	−2 per cent
Protein-bound iodine	6 μg. per cent
Cholesterol	272 mg. per cent
Fasting blood sugar	86 mg. per cent
Two hour postprandial sugar	75 mg. per cent
Urinary gonadotropins	6 mouse units
Urinary prolactin*	135 I.U.
17-Ketosteroids (before treatment)	21–26 mg. per 24 hr. (normal 5–15 mg. per 24 hr.)
17-Hydroxycorticosteroids (before treatment)	11–25 mg. per 24 hr. (normal 5–15 mg. per 24 hr.)
Vaginal smear	Hypoestrogenic pattern
Breast secretion	Fat globules
Skull film	Primary sellar tumor
Presacral air studies	Normal

* The urinary prolactin assay was secured through the kindness of Dr. Albert Segaloff.

further evidence of the presence of a pituitary lesion interfering with the secretion of gonadotropin.

Two explanations are possible for the occurrence of persistent lactation in patients with pituitary tumors. First there may be, as Forbes et al. have suggested, pressure on normally functioning secretory cells with resulting decreased release of the normal inhibitors of lactation. Second there may be production by the tumor of lactogenic and lactogenic supporting hormones. We believe that this latter possibility is more likely.

There is some reluctance to attribute hormonal production to the pituitary tumors of this syndrome because in most cases chromophobe tumors, the cells of which for the most part lack secretory granules, have been found. However, the experimental work of Furth [*7*] indicates that this criticism may not be relevant since he has been able to produce both dependent and autonomous mammatropic pituitary tumors composed largely of chromophobe cells in estrogen-treated rats.

The mammary glands of animals bearing these tumors have shown extensive hyperplasia of the ducts, distention of the alveoli with a milk-like secretion, and many mitotic figures. The liver, kidney, spleen and adrenal cortex of these animals were all greatly enlarged, suggesting a somatotropic as well as mammotropic effect of these estrogen-produced pituitary tumors.

Furth believes that the development of these mammotropic pituitary tumors depends upon hyperestrogenism. This theory would be compatible with the development of pituitary tumors and persistent lactation following pregnancy, the Chiari-Frommel syndrome. Since there is no suggestion either in our case and or in others not preceded by pregnancy that hyperestrogenism ever occurred, another explanation or a modification of Furth's theory is necessary.

The hormonal factors required for normal lactation are too complex for adequate discussion here. Although almost every hormone in the body may influence lactation, the two most important hormones would seem to be (1) the lactogenic or mammotropic hormone already briefly discussed, and (2) the corticosteroids. Ample experimental evidence has demonstrated the almost absolute dependency of lactation upon the presence of corticosteroids [*8*]. The exact part they play in the process of lactation is unknown. However, Folley's [*9*] concept that mammary tissue is preconditioned by corticosteroids so that it is able to respond to prolactin is a most attractive theory. It also has been shown that androgenic as well as hydroxycorticosteroids may participate in this action [*10*]. Thus the elevation of plasma 17-hydroxycorticosteroids found in the second and third trimester of pregnancy may play an important function in the preparation of breast tissue for lactation [*11,12*].

In view of the participation of adrenal cortical hormones in normal lactation, it seems important to consider how frequently hyperadrenal corticism is present in the syndrome of pathologic lactation.

Although in the majority of reported cases of persistent lactation measurements of urinary steroids have not been made, there has been a great deal of clinical evidence of increased adrenocortical function. Most of these patients have been obese and hirsutism has been common, suggesting some of the features of Cushing's syndrome. In addition, a case, with the classic features of the Chiari-Frommel syndrome has been reported in which a basophile adenoma was found, a pituitary lesion so often associated with Cushing's syndrome [*6*]. Although our patient lacked some of the classic features of the Cushing's syndrome, a round face, acne, truncal obesity and mild hirsutism were present. The clinical evidence of mild hyperadrenalcorticism is compatible with the moderately elevated steroid values of this patient. Interesting, too, is the frequent finding of large breast development in patients with Cushing's syndrome.

The occurrence of moderate increases in urinary steroid excretion in our case, 17-ketosteroids from 15 to 30 mg. per twenty-four hours and 17-hydroxycorticosteroids up to 25 mg. per twenty-four hours, and the increased urinary 17-ketosteroids found in all but two of the fifteen cases reported by Forbes et al. [*4*] strongly suggests that increased urinary levels of steroids may frequently be part of the syndrome of abnormal lactation.

The use of androgens and/or estrogens in previous similar cases to inhibit abnormal lactation has been only rarely effective. The success of radiation therapy to the pituitary in stopping lactation and returning steroid levels to normal in this case suggests that radiation therapy to the pituitary in adequate dosage should be tried in most cases of abnormal lactation. It would also seem to be indicated in those

cases in which the sella turcica appears to be normal radiographically.

SUMMARY

A case of persistent lactation associated with a pituitary tumor and increased urinary excretion of 17-ketosteroids and 17-hydroxycorticosteroids is presented. Cessation of lactation and a return to normal steroid excretion was accomplished by radiation therapy to the pituitary.

The possible relationship of persistent lactation to pituitary tumors, to prolactin or a mammotropic hormone, and to adrenalcortical function is discussed.

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Idiopathic Occlusive Disease of Digital Arteries with Associated Subacute Glomerulonephritis*

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THE following case report is that of a woman who entered the hospital because of gangrene of the fingers and toes, and subsequently died with a hemorrhagic pneumonitis. At autopsy, subacute glomerulonephritis was noted, and the gangrene proved to be due to a lesion of the digital arteries characterized by intimal and medial proliferation with obliteration of lumina and thrombosis. Clinical and pathological studies failed to reveal a specific cause for the arterial lesion.

This is the first case to be reported of idiopathic occlusive disease of digital arteries in association with glomerulonephritis. Following the case report and pathological findings are comments on the differential diagnosis of digital gangrene, in which the possible etiological factors in this case are considered.

CASE REPORT

A forty year old Hawaiian-Chinese housewife was admitted to The Queen's Hospital on August 26, 1957, complaining of pain and discoloration in the tips of several fingers and a toe, increasing over a three-week period. Initially there was only painless, yellowish discoloration in the pulp of the left fifth fingertip. Three days later the fingertip became black and the area of involvement enlarged. Subsequently other digits became involved and painful. There appeared to be pus at the junctions of necrotic and normal tissue. There had been no similar episode in the past; no blanching, pain or numbness in the digits prior to the black discoloration; no intermittent claudication or muscular cramps; and no ingestion of drugs, metals or ergot-containing foods.

Carious and missing teeth led the patient to a diet consisting mainly of fish, poi, soups and, rarely, meats. There was a cough productive of yellow sputum, but no hemoptysis, chest pain or feverishness. Menstrual periods over the preceding eighteen months had been infrequent, occurring every thirty-five to fifty days and lasting for two to three days, with scanty flow. The review of systems otherwise revealed no abnormalities.

Past medical history included presumptive minimal pulmonary tuberculosis with cervical and axillary adenitis, not confirmed by laboratory procedures, in 1936 to 1937. There was pneumonitis of the left lung in 1942 and bronchopneumonia in 1952. The latter illness was believed to have been complicated by congestive heart failure, although there had never been peripheral edema, or any subsequent dyspnea or orthopnea. Appendectomy was performed in 1952, and sterilization in 1953. A furuncle of the left breast continued to drain after incision-and-drainage procedure in April 1957. The patient had given birth to eleven children and had had two miscarriages. Ten children were alive and well. The cause of death of the eleventh at ten months of age was not known.

There was no history of diabetes, rheumatic fever, hypertension, renal disease or peptic ulcer.

Physical examination revealed a well developed, cooperative, middle-aged woman, complaining of pain in her fingers and toes. There was gangrene of the tips of the second, fourth and fifth digits of the left hand, the second and fifth digits of the right hand, and the fourth digit of the left foot. The involved digits were puffy and there were collections of pus at the several demarcation zones. (Fig. 1.) There were strong pulsations in radial, ulnar, posterior tibial and dorsalis pedis arteries bilaterally. The Allen test for patency of ulnar and radial arteries was normal. The hands and feet were of normal or perhaps slightly increased warmth, and were normal in color. There were no varicose veins, joint deformities, or scleroderma, and no acrocyanosis or gangrene of the ear lobes or nose tip.

Eyes, including fundi, were normal. Many teeth were missing, several were broken and the remainder had caries. The gums were inflamed, scarred and retracted. The thyroid was not enlarged. There were small surgical scars in the right side of the neck and right axilla. There were two small ulcerations medial

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to the left nipple, with colorless discharge. Coarse rales were heard in the left lung base posteriorly. The heart was not enlarged; the rhythm was regular at 88 per minute, and sounds were of normal character. There was a soft systolic murmur at the apex. Blood pressure was 90/60 mm. Hg. The abdomen was soft and diffusely tender to deep palpation. Liver, spleen and kidneys were not palpable. There was a surgical scar in the right lower quadrant of the abdomen. Pelvic examination revealed numerous nodules resembling sebaceous cysts on the vulva and a parous introitus. The cervix had anterior and posterior erosions. The uterus was small, in mid-position and movable. No adnexal masses were felt. Rectal and neurological examinations revealed no abnormalities.

Laboratory data revealed a red blood cell count of 3.98 million/cu. mm.; hemoglobin, 10.2 gm. per cent; hematocrit, 35 per cent; white blood cells, 17,600 per cu. mm., with 2 stabs, 81 mature neutrophils, 15 lymphocytes and 2 eosinophils per 100 cells. The red cells showed slight hypochromia and anisocytosis. The platelet count was 260,000/cu. mm. These counts were essentially unchanged one week after admission. The urine was cloudy amber in color with a pH of 6, specific gravity 1.012, protein 2-plus, no sugar or acetone, 15 to 20 white cells and 8 to 10 red cells per high power field, and a few coarse and fine granular casts per low power field. The specimen was sterile. Blood culture was negative. Discharge from the left breast sinus contained Proteus mirabilis overgrowing Micrococcus pyogenes var. albus and diphtheroids. The sputum contained abundant Hemophilus influenzae, moderate Neisseria catarrhalis, a few pneumococci and rare Pseudomonas aeruginosa. Sputum smear and culture were negative for Mycobacterium tuberculosis.

Fasting blood sugar was 97 mg. per cent; serum uric acid, 6.3 mg. per cent; blood urea nitrogen, 13.5 mg. per cent; cholesterol, 183 mg. per cent, with 65 per cent esters; Kunkel (zinc sulfate), 18.7 turbidity units; alkaline phosphatase, 1.3 Bodansky units; phosphorus, 3.4 mEq./L.; calcium, 4.8 mEq./L.; protein-bound iodine, 5.4 γ per cent; total serum proteins, 6.5 gm. per cent, with albumin, 3.3, and globulin, 3.2; gamma globulins, 2.2 gm. per cent by chemical analysis. By serum protein electrophoresis, the albumin was 30.3 per cent of the total protein (2.06 gm. per cent); α 1 globulin, 8.9 per cent (0.60 gm. per cent); α 2 globulin, 14.8 per cent (1.01 gm. per cent); β globulin, 20 per cent (1.36 gm. per cent), and γ globulin, 26 per cent (1.77 gm. per cent). The blood type was O, Rh positive.

No L.E. cells were seen in several examinations by three different technics. No cryoglobulins were detected. The serologic test for syphilis was negative. Clot retraction was normal. Several determinations for urinary porphobilinogen were negative.

The electrocardiogram demonstrated sinus tachycardia, clockwise rotation of the heart and non-specific T wave changes. X-ray examination of the chest demonstrated distortion of the bronchovascular markings in the lower lobe of the left lung, with an associated peribronchial infiltration. The heart was not enlarged.

FIG. 1. Gangrene of the tips of several fingers. A similar lesion was present on one toe.

X-ray studies of the hands and feet revealed small notch-like defects in the subarticular cortical aspects of the heads of multiple middle phalanges of the right and left hand. There was no apparent recent osteomyelitis or periosteal reaction. Flat plate of the abdomen revealed that both kidneys were normal in contour and position, the spleen was not enlarged, and there were no apparent unusual calcific deposits in the walls of the abdominal aorta or its iliac branches.

The patient was placed at bedrest and treated with Priscoline,® 25 mg. orally every four hours and procaine penicillin administered intramuscularly; heat was applied to the abdomen. Two days after admission the gangrene was extending in the right second finger and the left fourth toe, and the right fourth finger had become dusky in color. After another two days, the duskiness had cleared from the right fourth finger, but the tip of the right third finger had become gangrenous. Nine days after admission, the gangrene was worse in the right second and third fingers and in the left fourth toe. Small areas appeared to be clearing on the right fifth finger. The left fingers were unchanged. Priscoline was given intramuscularly, 5 mg. every four hours.

One week after admission, the patient began to complain of nausea, substernal burning sensation and vomiting. She was given a peptic ulcer diet. On the ninth hospital day an upper gastrointestinal series was performed because of persistent symptoms. In the lesser curvature of the gastric antrum or adjacent pars media was a small ulcer crater; a constant irregular filling defect was also present along the lesser curvature, extending to the pylorus, which was rigid. These findings were regarded as indicative of an extensive ulcerative type of gastric malignancy. The duodenal bulb was not clearly visualized, but the duodenal

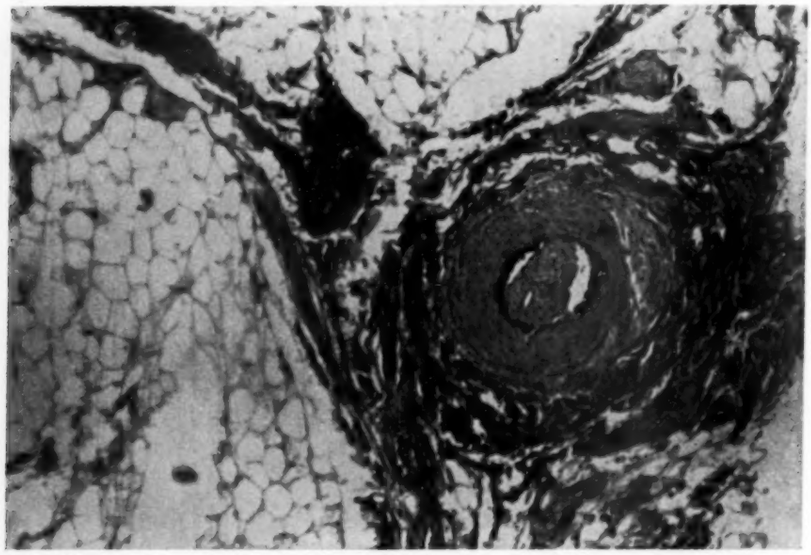

FIG. 2. Elastic tissue stain of digit, demonstrating proliferation of media and intima with thrombosis of digital artery.

loop appeared to be normal. There was also a large mid-esophageal traction diverticulum, with no evidence of esophageal obstruction. X-ray film of the chest at this time revealed no active pulmonary or pleural lesions. The heart and diaphragm appeared normal. There was occult blood in the feces. No free acid was found in fasting gastric juice. Histamine elicited 10 degrees of free acid.

On the eighteenth hospital day laparotomy was performed. A gastric ulcer in the lesser curvature, hypertrophic pylorus and pylorospasm were noted. A subtotal gastric resection was carried out with a Billroth I anastomosis. Liver biopsy was performed and the sinus of the left breast was excised. Blood loss during surgery was estimated at 400 cc., and 1,000 cc. of whole bank blood and 1,500 cc. of intravenous fluids were given.

Microscopic examination of tissues removed at surgery revealed the following: The findings in the stomach were those of a benign gastric ulcer. Most blood vessels appeared normal, except for the occasional presence of leukocytes in the walls, possibly due to diapedesis. In some medium-sized arterioles there was fibrosis of the walls with narrowing of the lumina and infiltration of lymphocytes and polymorphonuclear leukocytes. In the liver, there was slight-to-moderate periportal infiltration, consisting mainly of eosinophils, plasma cells and monocytes. The hepatic cells appeared normal and no other changes were noted. In the breast, only chronic inflammatory changes were noted.

The patient had a rather uneventful postoperative course, except for a respiratory infection on the sixth postoperative day. It was characterized by nasal discharge and a cough productive of small amounts of mucopurulent sputum. There was no fever and the lungs were clear on physical and x-ray examination. The symptoms subsided and the patient was discharged on the fifteenth postoperative day. The gangrenous digital lesions had remained stationary.

The patient was seen in the outpatient department three days after discharge from the hospital. She had daily bowel movements and was eating well, although she had a feeling of fullness after eating small amounts. There was a productive cough, but the lungs were clear on physical examination. The patient was given an antitussive, and therapy with ferrous sulfate was begun. She was to be seen again two weeks later. The finger and toe lesions were unchanged.

On October 10, 1957, sixteen days after discharge and about six weeks after her initial admission to the hospital, the patient had sudden hemoptysis at home and died. Autopsy was performed.

At autopsy the fingers and toes, on gross appearance, were much the same as described previously. Microscopically, keratin elements were seen splitting away from the underlying normal epidermis. Within the dermis, there was a moderate generalized infiltrate, increased focally, and consisting largely of lymphocytes and plasma cells. A focus of granulomatous response was present in the dermis in one section. Cells had proliferated in the arterial walls, partially occluding the lumina. In one section, the arterial proliferation appeared focal or nodular, confined to one part of the circumference. Onion-peeling of an arteriole, with obliteration of the lumen, was seen. No perivascular inflammatory infiltrate was apparent. Elastic tissue stains (Fig. 2) showed that in some arteries the proliferation was largely intimal, in others medial, and in some both types of proliferation were present. Thrombi occluded some vessels.

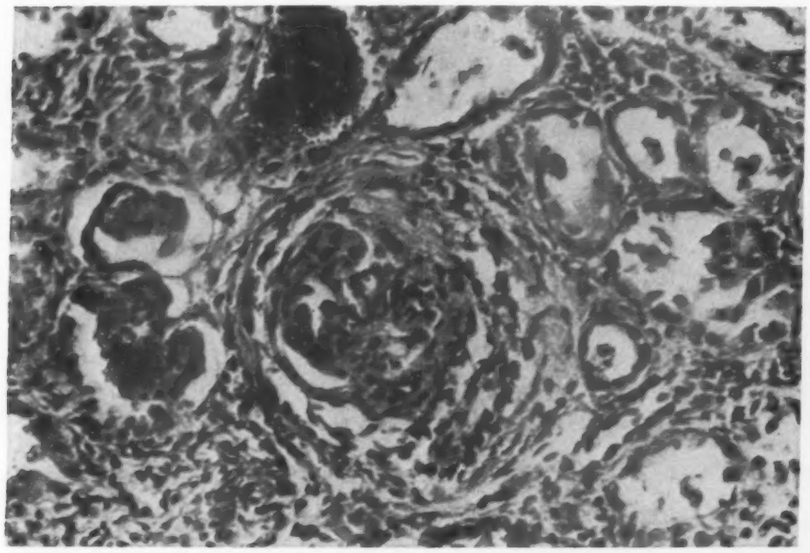

FIG. 3. Section of kidney demonstrating the typical glomerular lesion of subacute glomerulonephritis.

About 200 cc. of cloudy fluid was present in either pleural cavity, and the pleurae were smooth and glistening. The lungs were extremely red and firm throughout. Excessive edema fluid and blood poured from the cut surface. The right lung weighed 1,100 gm., the left 800 gm. The bronchi were slightly congested and filled with bloody froth. Peribronchial lymph nodes were firm and gray. Microscopically, alveoli were filled with erythrocytes, and in some, leukocytes, principally hemosiderin-containing macrophages, were prominent. Eosinophilic exudate was also present. In some areas alveolar septums were widened and showed hyaline degeneration. Breaks in continuity of septums were seen. Within the septums there was extensive mixed infiltrate, including polymorphonuclear leukocytes. The walls of small arteries were thickened, often showing hyalinization, and there was frequently a periarterial mixed infiltrate. The bronchioles were relatively free of cells, except for erythrocytes, although one was loaded with acute inflammatory cells. There was mixed peribronchial infiltrate, consisting mainly of polymorphonuclear leukocytes and monocytes. Focal fibrosis of the parenchyma was noted. The pleura was thickened and contained an infiltrate, mainly polymorphonuclear leukocytes, with a few plasma cells and monocytes.

About 100 cc. of clear fluid lay within the pericardial sac. The heart weighed 325 gm. Cardiac valvular measurements were within normal limits. Slight atherosclerosis of the coronary arteries was present. On microscopic examination there was focal atrophy of myofibrils, with only the connective tissue stroma persisting. In such areas, a few inflammatory cells were seen, mostly small mononuclear cells. Elsewhere, there was focal mixed infiltrate lying between muscle fibers and consisting primarily of neutrophils, lymphocytes and plasma cells. In perivascular tissue about the small arteries, there was mixed infiltrate.

Grossly, the line of anastomosis between the cardia of the stomach and the duodenum appeared to be intact. Microscopically, suture material was surrounded by focal granulomatous reaction, including many plasma cells and macrophages. Throughout the gastrointestinal tract there was intermittent congestion of mucosal vessels. There were six ulcers in the ileum, about 10 cm. apart. On gross examination these ulcers had firm bases. Microscopically, there was loss of mucosal glands and, in their place was fibrin with entrapped and degenerating leukocytes. Lymphocytes lay between muscle fibers and were scattered throughout the tissue, not localized about vessels. Hyalinization had occurred in the walls of some vessels. Grossly, there was also an ulceration of the sigmoid mucosa, about 1 cm. in diameter.

On gross examination the liver was pale but otherwise normal. Microscopically, periportal infiltrate, mainly mononuclear, was fairly prominent. Portal arterioles had thickened, hyalinized walls. Slight portal fibrosis had occurred. Sinusoids were dilated and inflammatory cells lay within them. Central veins were likewise dilated and some were surrounded by fibrosis. Individual hepatic cells appeared necrotic, with loss or degeneration of nuclei, and other hepatic cells had large or double nuclei or were mitotic. Focal fatty metamorphosis was seen.

The kidneys weighed 200 gm. each. The capsules stripped with moderate ease revealing smooth surfaces. The kidneys were extremely pale and bulged from the cut surfaces. Creamy material was present within the pelves of both kidneys. There was no accentuation of the corticomedullary line. Microscopically, adhe-

sions forming fibrous crescents lay across Bowman's spaces. (Fig. 3.) All glomeruli were involved to varying degrees, some being completely fibrosed and hyalinized, others showing focal hyalinization of the capillary loops. Arteriolar walls were proliferated and thickened, with focal hyalinization. In one, a thrombus partly occluded the lumen. Periarterial infiltrate was insignificant. Hyaline degeneration and swelling of some tubular cells were seen. Within some tubules there were hyaline and reddish granular casts. Mononuclear cells were scattered throughout the stroma. One section through the renal pyramid contained extensive acute inflammatory infiltrate, as well as focal hemorrhage. Tubules were extensively damaged in this section.

The adrenal cortices were pale. Microscopically, adenomatous formations were present in the *zona fasciculata*.

Spleen, brain and vertebral marrow were grossly and microscopically within normal limits.

The final anatomical diagnoses were: subacute glomerulonephritis; hemorrhagic pneumonitis, massive; intimal and medial thickening of small digital arteries with some thromboses; gangrene of fingers and toes; myocarditis, slight; chronic passive congestion of liver, slight; portal cirrhosis, mild; ulcerations of the mucosa of the ileum, multiple, acute; pericardial effusion, 100 cc.; and pleural effusion, bilateral, 200 cc. either side.

COMMENTS

The site of pathologic changes in this patient's digits was thought to be the smaller digital arteries, since clinical examinations attested to the patency and adequacy of the medium-sized vessels of the extremities. It was further believed that organic changes were present to account for the progressive gangrene. The question arose as to whether the organic changes were primary, or were perhaps secondary to vasospasm, since it has been reported that long-standing arteriospasm has been followed by organic occlusion of the vessels [*1*].

Arterial spasm can be a part of Raynaud's disease, or associated with hypoglycemia, hypothyroidism, intoxication with ergot, heavy metals or certain drugs, or neurologic disorders such as lesions in the mid-brain, internal capsule, spinal cord, or a postsympathectomy state. However, the patient did not have Raynaud's phenomenon: The symptoms were of relatively short duration and lacked intermittency. Nor was there any history of ingestion of toxic agents, or any findings to suggest the other causes of vasospasm listed above. It was therefore thought that a primary organic occlusive disease of the smaller arteries was present in this case, and attempts were made to identify the exact type of occlusive disease clinically. The differential diagnosis included: multiple emboli, scleroderma, disseminated lupus erythematosus, rheumatoid arthritis, polyarteritis nodosa, sensitization angiitides, polycythemia vera, cold hemagglutination, cryoglobulinemia, cold injury, ball valve thrombus in the left auricle, leprosy, thrombotic thrombocytopenic purpura, syphilis, porphyria, and certain other specific infectious processes. All of these conditions were excluded on clinical grounds and with appropriate laboratory studies, so that the antemortem diagnosis was idiopathic occlusive disease of small arteries.

Postmortem findings did not help to identify the arterial lesions in this patient definitely with any previously described specific lesions. Similarities were noted, however, between our case and others reported previously under such diagnoses as endarteritis obliterans [*2*], periarteritis nodosa [*3*], and disseminated lupus erythematosus [*4*]. These will be described.

Endarteritis obliterans of digital arteries is a relatively uncommon disorder [*2*], characterized by intimal proliferation of smaller arteries, leading to trophic disturbances of the soft parts of the digits. One or more digits may be involved, in upper or lower extremities, unilaterally or bilaterally. The condition is more common in the upper extremities. Various toxic, irritative, infectious and sensitization causes have been considered in the etiology, but none proved. It is quite possible that this condition is caused by many factors, as yet unknown. The diagnosis is made from the clinical picture, with ulcerative or gangrenous lesions, in the presence of good pulses in the medium-sized arteries and normal results of tests for circulation in these medium and larger-sized vessels, and by exclusion of known causes of occlusion of smaller-sized arteries. The prognosis is stated to be fairly good, with gangrene usually self-limited. Occasionally the process may extend rapidly along a vessel, leading to extensive gangrene, and is then referred to as endarteritis proliferans. Sixty per cent of cases occur during the fourth, fifth and sixth decades. Men are affected more frequently than women, and Caucasians more frequently than Negroes. Presenting symptoms include pain, coldness, numbness, cramps, burning sensations and fatigue.

The present case would fit into the category

of endarteritis obliterans, were it not for the added feature of medial proliferation, in addition to the expected intimal proliferation.

Periarteritis nodosa and its relationship to other angiitides has been reviewed thoroughly and critically by Zeek in a series of articles [*5–7*]. Zeek employs the generic term necrotizing angiitis in referring to lesions of arteries and veins, which, in their fully developed stage, are characterized by fibrinoid necrosis and inflammation involving all three layers of vessel walls. Among the necrotizing angiitides are: (1) hypersensitivity angiitis; (2) allergic granulomatous angiitis; (3) rheumatic arteritis; (4) periarteritis nodosa; and (5) temporal arteritis. There are probably other unclassified types. Excluded from Zeek's classification are arteriolar necrosis of malignant hypertension (little or no inflammatory reaction) and infectious endarteritis or periarteritis (no fibrinoid necrosis).

We believe that the present case, presenting no significant degree of inflammation in vessel walls and no fibrinoid necrosis, should not be considered among the necrotizing angiitides.

Case III reported by Meacham and Weisberger [*4*] involves Raynaud's phenomenon as one of the initial less common manifestations of disseminated lupus erythematosus. Pathologic diagnosis at the time of amputation of fingers for pain and cyanosis was proliferative endarteritis and organizing thromboses of arteries, etiology undetermined. Subsequent biopsy specimen of the skin in the same patient revealed widespread arteriolar thrombi and focal chronic arteriolitis. More typical findings of disseminated lupus erythematosus later developed in the patient.

Our patient had findings similar to those described at the time of amputation in the case just referred to, but in the present patient results of several L.E. preparations were negative, and postmortem examination of other tissues revealed nothing to suggest disseminated lupus erythematosus.

Glomerulonephritis has not previously been reported, to the best of our knowledge, in association with occlusive disease of peripheral arteries, except that there is an increased tendency towards thromboses in the nephrotic stage. We must therefore either assume that the coincidence is fortuitous, or else that there may be some underlying rare etiologic condition common to both diseases in our patient. We have considered some of the diffuse mesenchymal disorders as perhaps explaining the entire clinical picture. However, with each instance we have arrived at certain inconsistencies.

The terminal event in this patient's life was undoubtedly the hemorrhagic pneumonitis. The microscopic findings were consistent with either a viral pneumonitis or with a so-called antigenic pneumonitis such as is seen in uremic states. The 1957 epidemic of Asian influenza was present in Hawaii at the time the patient died, and there were other cases of fatal hemorrhagic pneumonitis thought to be due to the influenza virus. There have been other reported cases of hemorrhagic pneumonitis in Asian influenza [*8*]. It is quite possible that this indeed was the cause of death in the present case. However, the microscopic findings in the kidneys make a uremic state possible. Hemorrhagic pneumonitis has been described also in association with the renal lesions of periarteritis nodosa and acute glomerulonephritis [*9*], but, as mentioned previously, there were no findings in this patient to suggest a diagnosis of periarteritis nodosa.

The possibility exists that there was an underlying hypersensitivity state present in this patient. Certainly, glomerulonephritis has been considered to be a result of sensitivity reactions [*10*]. However, the arterial lesions in this patient had neither of the major characteristics of hypersensitivity angiitis, fibrinoid degeneration or inflammatory changes of marked degree.

SUMMARY

A case is reported of idiopathic occlusive disease of digital arteries in association with subacute glomerulonephritis, a hitherto unreported combination. The differential diagnosis of digital gangrene is discussed, and similarities are noted between certain findings in this case and in cases of endarteritis obliterans, periarteritis nodosa, and disseminated lupus erythematosus. The possibility is entertained that an underlying hypersensitivity state existed in this patient.

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Lobar Pneumonia with Bacteremia Caused by Bacterium Anitratum*

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The recent literature has revealed repeated references concerning the changing relationships between chemotherapeutic agents and known bacteria. Emergence of mutant bacterial strains and selective overgrowth of resistant strains are factors which have received progressively increasing attention. This paper may illustrate a third and less commonly discussed factor of bacterial resistance, that is, the conversion of a bacterium from a saprophyte to a pathogen. The basis for this article is a case of severe pneumonia and accompanying Bacterium anitratum bacteremia.

A brief review of the English literature failed to reveal a similar case reported in detail and, indeed, emphasized the fact that very little is yet known of this organism. Early workers concentrated on Diplococcus mucosus, a gram-positive coccus. Not until McFarlan [*1*] in 1941, and Stuart and associates [*2*] in 1949, were attempts made to cull and classify the previous literature. In their classification Stuart and his associates included a gram-negative organism which they named B5W. This is similar to if not identical with B. anitratum, identified by Schaub and Hauber [*3*] in 1948. The sixth edition of Bergey's Manual [*4*] follows the catalogue of De Bord [*5*] and does not list B. anitratum. Brooks and Landers [*6*], and others, consider it to be a member of the family Parvobacteriaceae, tribe Mimeae, which at this time includes three species: Mima, Herellea and Colloides. Some investigators consider B. anitratum to belong to the Herellea species, but this has yet to be generally accepted.

Bacteriology. B. anitratum is a large (0.8 μ.) gram-negative diplococcus which is encapsulated and has a pleomorphic nature. Specific strains may be either motile or non-motile. On agar it is characterized by large white or buff mucoid colonies. Biochemically it is capable of reducing only the simpler carbohydrates of five carbons or less. Because of its inability to reduce nitrates, it was christened anitratum. This organism, as well as all members of the Mimeae tribe, mimic Neisseria gonorrhoeae to such a degree that differentiation on smears is impossible. The consequent hazard of diagnosis by smear in genitourinary tract disease has been pointed out by Ewing [*7*] and De Bord [*8*].

Pathogenicity. The fact that B. anitratum has been infrequently reported in the past bespeaks its low pathogenicity. The majority of workers consider it to be an unusual saprophyte. However, the more recent literature mentions accounts of human disease, often with a fulminating fatal ending. De Bord [*8*] reported forty-five isolations of Mimeae from conjunctival and vaginal cultures. Brooke [*9*], Waage [*10*] and others, have reported approximately twenty-four infections of the central nervous system. The most frequent source of isolation, approximately 189 times, was from the genitourinary tract, as shown by Deacon [*11*], and others. Less frequent sources were: blood culture, skin wounds, sputum, throat, abscesses, feces, pleural fluid, pericardial fluid, gastric contents, cellulitis, eye, ear, prostate, uterus, bile, lungs, and aspiration material at bronchoscopy, reported by many authors.

We were able to find only four reports of pneumonic infection with B. anitratum: Schaub and Hauber [*3*], Ferguson and Roberts [*12*], Simpson and Crossley [*13*], and Faust and Hood [*14*]. In none of these cases did the patient survive. Only four patients having serious illness and from whom a Mima organism was isolated were recorded as remaining alive: three with

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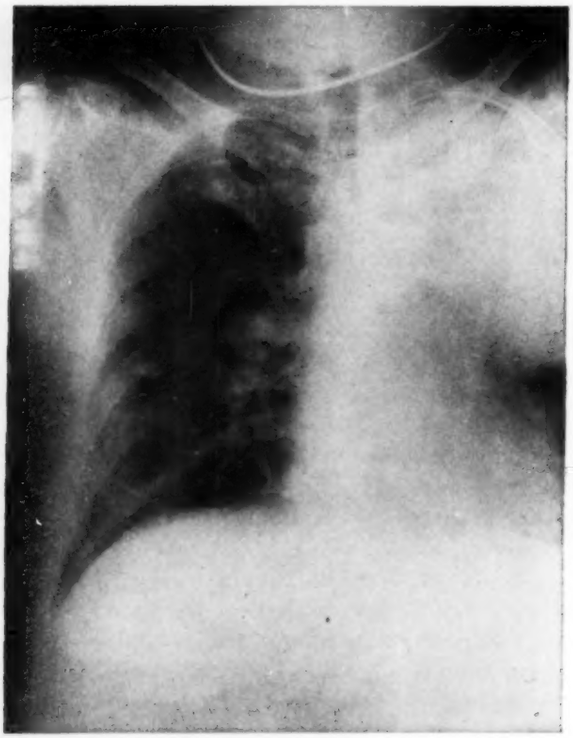

FIG. 1. Roentgenogram showing an encapsulated empyema.

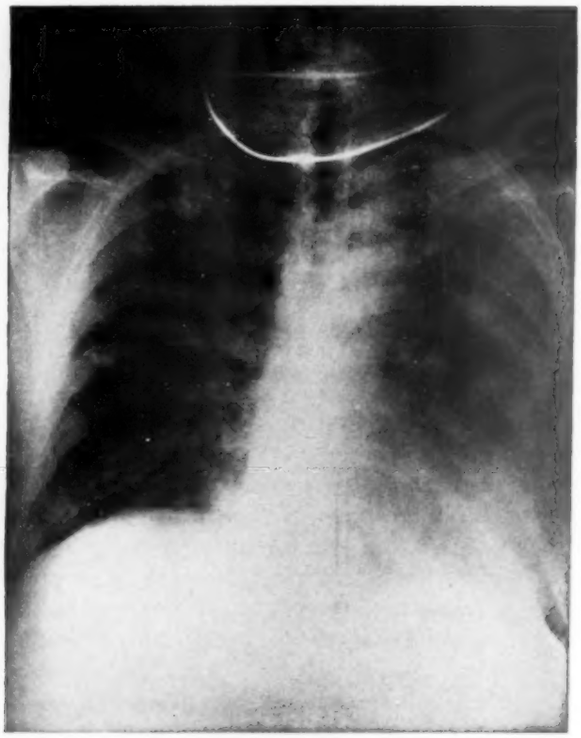

FIG. 2. Second roentgenogram twenty-four hours later showing diffuse involvement of both lungs.

meningitis treated with sulfonamides, as reported by McFarlan [*1*], De Bord [*15*] and Townsend and associates [*16*], and one with subacute bacterial endocarditis given oxytetracycline, as reported by Pike and associates [*17*].

CASE REPORT

A fifty year old man was admitted to the hospital because of pain in the chest, nausea and vomiting of twenty-four hours' duration. Several months previously he had noted substernal pains of short duration brought on by exertion. On one occasion, two weeks prior to admission, he had experienced a single episode of similar nature lasting six hours and associated with numbness in both arms and hands. In the interim he had continued his normal activities despite increasing fatigability. After breakfast the day before admission he became nauseated and vomited. Shortly thereafter he experienced sharp pains in the left anterior and lateral thorax which were aggravated by deep inspiration.

On admission the oral temperature was 100.8°F., and respirations 36. A few moist rales were noted in both bases posteriorly, and on deep inspiration a pleural friction rub was heard, predominantly in the left apex posteriorly. Except for a heart rate of 96, there were no abnormal cardiac findings. Moderate tenderness with muscle guarding was present in the right upper quadrant of the abdomen, and the liver was palpated 6 cm. below the right costal margin. No peripheral edema was observed.

The patient's condition deteriorated rapidly. The temperature remained in the vicinity of 102°F., pulse rate 140, and respirations 40. He became extremely toxic and disoriented, and persistent cyanosis developed despite continuous oxygen therapy. Morphine sulfate, digitalis, penicillin and dihydrostreptomycin were given.

Roentgenographic examination of the chest showed an infiltration in the upper lobe of the left lung consistent with an encapsulated empyema or a lobar pneumonia. New roentgenograms (Figs. 1 and 2) taken twenty-four hours later revealed diffuse involvement of both lungs which was interpreted as pulmonary edema, and conformed with the clinical findings. Initially the hemoglobin was 13.2 gm. per cent; white blood cell count, 17,750 per cu. mm.; and a differential count of 81 per cent polymorphonuclears, 9 per cent non-segmented polymorphonuclears, 6 per cent lymphocytes and 4 per cent monocytes. Urinalysis was within normal limits. Stool, urine and blood cultures were made. Serial electrocardiograms revealed only sinus tachycardia and transitory anoxia. Because the patient's clinical course was deteriorating, sulfasoxazole was added to the regimen.

By the third hospital day the physical findings of lobar consolidation in the upper lobe of the left lung

TABLE I
SPUTUM AND BLOOD STUDIES

Dates	Findings
	Sputum Cultures
11/26/55....	Yeast, Klebsiella
11/30/55....	Neisseria Non-hemolytic streptococci Staphylococci Yeast Klebsiella
12/1/55.....	Non-hemolytic streptococci, diplococci,
12/2/55.....	B. anitratum
12/5/55.....	B. anitratum
12/6/55.....	Yeast
12/7/55.....	Yeast, beta hemolytic streptococci
12/30/55....	Hemolytic staphylococcus albus (bronchial aspirations)
	Blood Cultures
11/26/55....	B. anitratum (24 hr.) (1 wk.) (2 wk.), B. anitratum (24 hr.) (1 wk.) (2 wk.), B. anitratum (24 hr.) (1 wk.) (2 wk.)
12/3/55.....	No growth (24 hr.) (1 wk.) (2 wk.)
12/5/55.....	No growth (24 hr.) (1 wk.) (2 wk.)
12/9/55.....	No growth (24 hr.) (1 wk.) (2 wk.)
12/10/55....	No growth (24 hr.) (1 wk.) (2 wk.)

TABLE II
SENSITIVITY STUDIES ON B. ANITRATUM*

Antibiotic	Blood (11/25/55)	Sputum (11/28/55)	Sputum (12/2/55)	Sputum (12/5/55)
Tetracycline	2	3	3	3
Oxytetracycline	2	3	3	3
Chlortetracycline	2	3	3	3
Triple sulfa	0	0	3	2
Erythromycin	0	0	3	2
Streptomycin	1	1	..	..
Penicillin	0	0	0	0
Chloramphenicol	3	3	3	3

* Filter paper disk method: 3, strongly sensitive; 2, moderately sensitive; 1, weakly sensitive; 0, resistant.

appeared, while a few basilar rales persisted. A cough developed, producing a rusty tenacious sputum. The friction rub persisted through the fourth hospital day, then disappeared without recurrence. The patient continued to be gravely ill for the first week, and became more cyanotic whenever oxygen was discontinued. Because he failed to respond to the previous management, administration of oxytetracycline and chloramphenicol was begun. Owing to continued evidence of cardiac failure, digitalization was augmented with cedilanid. Dyspnea and grunting respirations predominated. The productive cough continued.

During the second hospital week there was gradual

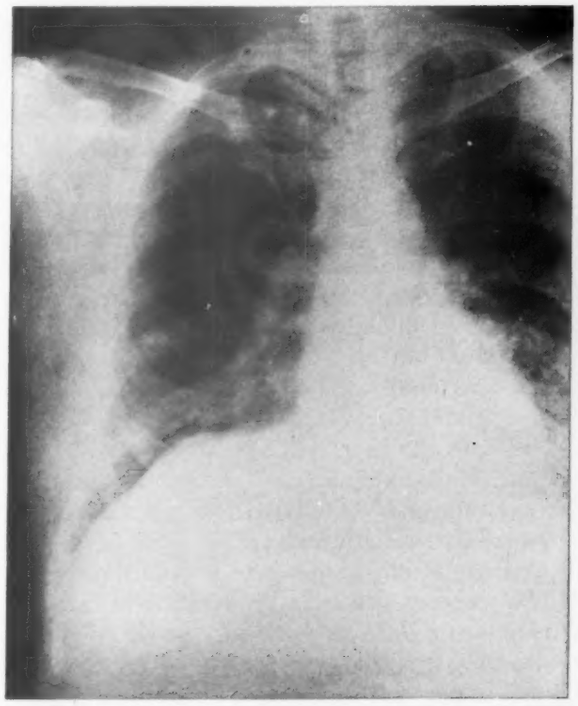

FIG. 3. Follow-up roentgenogram three years later showing clearing.

quiescence of physical signs and symptoms. The patient became less toxic and administration of oxygen was discontinued on the thirteenth hospital day. The cough subsided during this period. The roentgenographic picture of pulmonary edema remained relatively constant until the fourteenth hospital day, when fluid, suggesting a recurrence of an encapsulated empyema, was noted in the upper lobe of the left lung. Five days later some spotty clearing was noted. By the end of one month clearing was progressing but not yet complete, with evidence of extensive fibrosis and retraction.

The hemoglobin reached a low of 11.3 gm. per cent on the thirteenth hospital day but rose with convalescence to 12.8 gm. A maximum leukocytosis of 23,650 per cu. mm. was recorded on the thirteenth hospital day, but the count returned to normal rapidly. The polymorphonuclear elevation remitted more slowly. Serial urine and stool cultures were consistently negative. Six gastric aspirations were negative for acid-fast bacilli, although the Mantoux test was strongly positive in the first strength (1:10,000). Sputum and blood studies are shown in Table I.

After the second week the patient began to regain strength, and convalescence progressed slowly but uneventfully. A four month follow-up revealed complete clinical and laboratory recovery. Furthermore, repeated follow-up examinations over a three-year period have revealed the patient to be completely recovered. (Fig. 3.)

COMMENTS

As shown in Table II, initial sensitivity studies on the organism isolated from our patient's blood showed marked sensitivity to chloramphenicol. Only moderate sensitivity was demonstrated to tetracycline, oxytetracycline and chlortetracycline. Penicillin, triple sulfa (di-meth-mer) and streptomycin proved to have no inhibitory effect. It is interesting to note that after nine days of therapy, when the organism was again isolated, this time from the sputum, it was found to be more sensitive to tetracycline, oxytetracycline and chlortetracycline than in the initial blood studies.

Several antimicrobial agents were employed while awaiting sensitivity studies. Because the fever diminished in our patient by lysis and not by crisis, one could not argue strongly for the specificity of any of the antimicrobial agents.

The present case calls attention to the fact that unknown organisms may be encountered with increasing frequency as virulent pathogens. This is due in part to the widespread use of antibiotics which have tended to change the previous pathogenic flora.

SUMMARY

A severe case of lobar pneumonia and bacteremia caused by B. anitratum is presented, together with a review of the literature on this organism and its bacteriological characteristics.

Therapeutic experiences in general have shown the tetracyclines to be the most efficacious in treating infections caused by B. anitratum.

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Pulmonary Edema in Acute Heroin Poisoning*

Report of Four Cases

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PULMONARY edema has been described in a wide variety of conditions and has been the object of much interest and controversy [*1,2*]. Although some authors list opium poisoning among the clinical conditions associated with an abnormal extravascular water storage in the lungs [*2–4*] only a few accounts of the clinical and roentgenologic aspects of this entity can be found in the literature [*5–10*].

In the last three years at Bellevue Hospital we have encountered four instances of pulmonary edema in patients known to have had an overdose of heroin. It is the purpose of this report to describe the features of this condition, review the pertinent literature, and discuss possible pathophysiological mechanisms.

CASE REPORTS

CASE I. A. V., a twenty-four year old Puerto Rican man, on April 19, 1955 suddenly lost consciousness after an intravenous injection of heroin. He was comatose when brought to the emergency room of another hospital. Because of venipuncture marks and pin-point pupils, N-allylnormorphine (5 mg.) was given intravenously with some clearing of the sensorium, and the patient was transferred to this hospital. The patient had been taking heroin regularly for about one year. One month earlier he had discontinued its usage until the day of admission. †

Physical examination revealed a lethargic, cyanotic, orthopneic man who was raising scant whitish sputum. The respiratory rate was 24 per minute; temperature, 99°F., pulse rate, 88 per minute; and blood pressure, 140/80 mm. Hg. The pupils were miotic and did not react to light. There was dullness on percussion over both lung bases posteriorly. Fine rales were scattered throughout both lung fields. The remainder of the physical examination was noncontributory.

Urinalysis on admission showed 2 plus albumin, 3 plus sugar. The hemoglobin was 16.5 gm. per cent; white blood cell count, 3,150 cells per cu. mm.; and the differential was within normal limits. A roentgenogram of the chest taken on admission (Fig. 1A) revealed diffuse blotchy areas of increased density in the lower two-thirds of both lung fields. The hilar and paracardiac areas were most affected with relative clearness of the extreme periphery and apical areas. The cardiac outline was normal.

A diagnosis of bronchopneumonia was made, and penicillin therapy started. Within twelve hours the cough diminished, rales were no longer audible and there was marked subjective improvement. An x-ray film of the chest taken sixteen hours after admission (Fig. 1B) showed marked clearing of the pulmonary densities seen in the previous film. Two days after admission the patient felt well, and physical examination was within normal limits; glycosuria and albuminuria had subsided.

CASE II. L. D. V., a fifteen year old Puerto Rican boy, on June 28, 1957 suddenly became unconscious after an intravenous injection of heroin. On arrival at another hospital the patient was comatose and cyanotic. The pupils were miotic and failed to respond to light. He recovered consciousness after 10 mg. of N-allylnormorphine was given intravenously and was then transferred to this hospital. The patient, who had used heroin intermittently for one year, thought that this was a greater than his usual dose.

Physical examination revealed a tachypneic, apprehensive, orthopneic, cyanotic youth who had a hacking cough. His respiratory rate was 48 per minute; temperature, 99°F.; pulse rate, 144 per minute; and blood pressure, 100/40 mm. Hg. The pupils were miotic and did not react to light. There was flaring of the alae nasae. Rales and rhonchi were heard over the lower two-thirds of both lung fields. The extremities showed linear venipuncture scars; otherwise there were no significant findings on physical examination.

† We were unable to estimate the actual amount of heroin taken in this and subsequent cases because of the extreme variability in purity of the drug sold to addicts, as well as the unreliability of the histories given by these patients.

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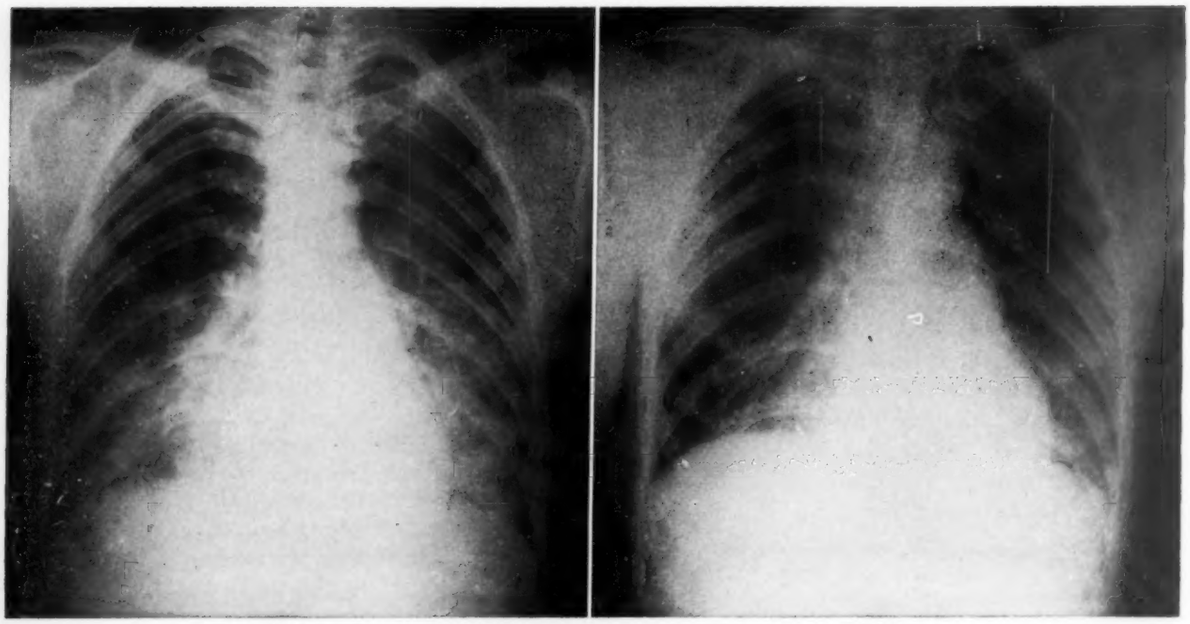
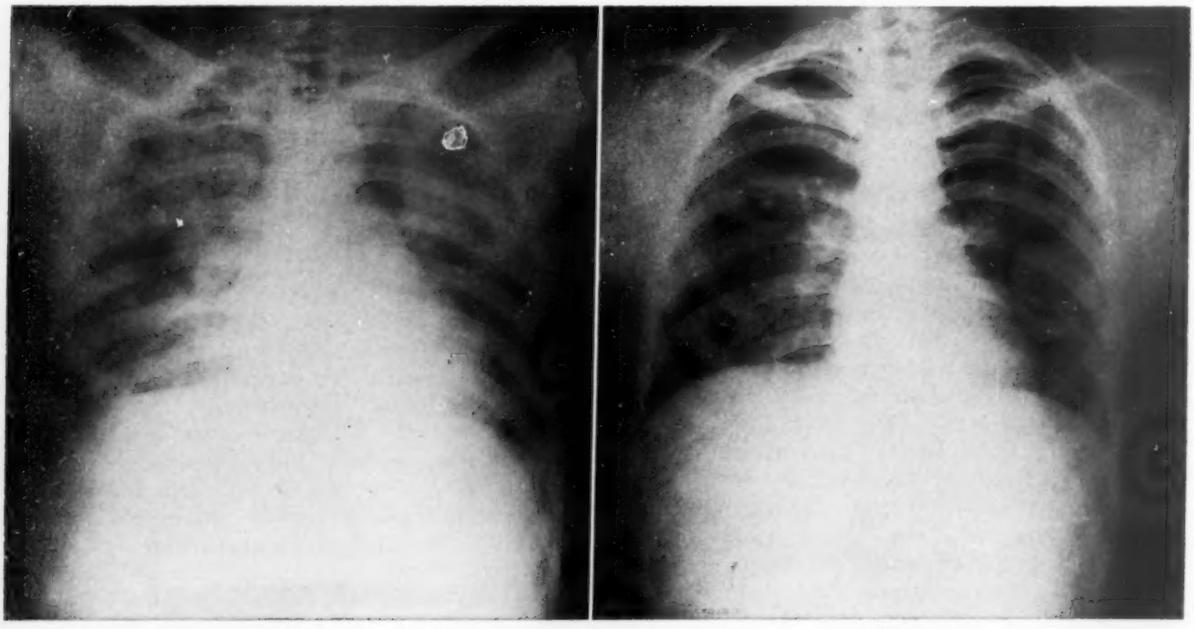


1A 1B

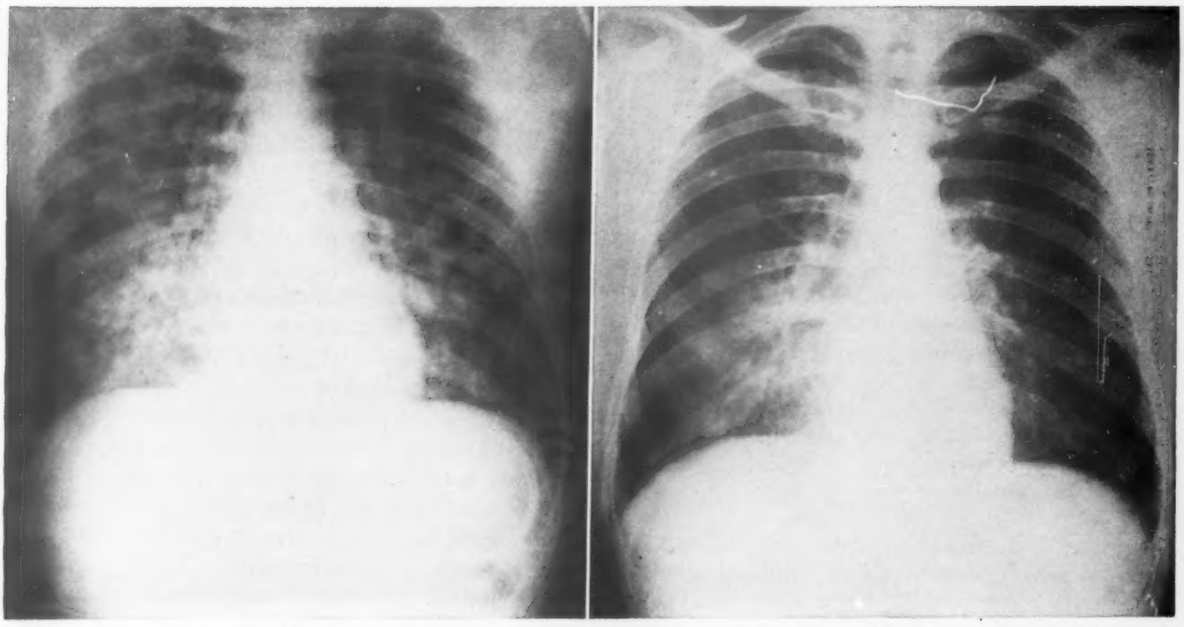
FIG. 1. Case I. A, chest roentgenogram taken on admission, B, follow-up examination sixteen hours later.



2A 2B

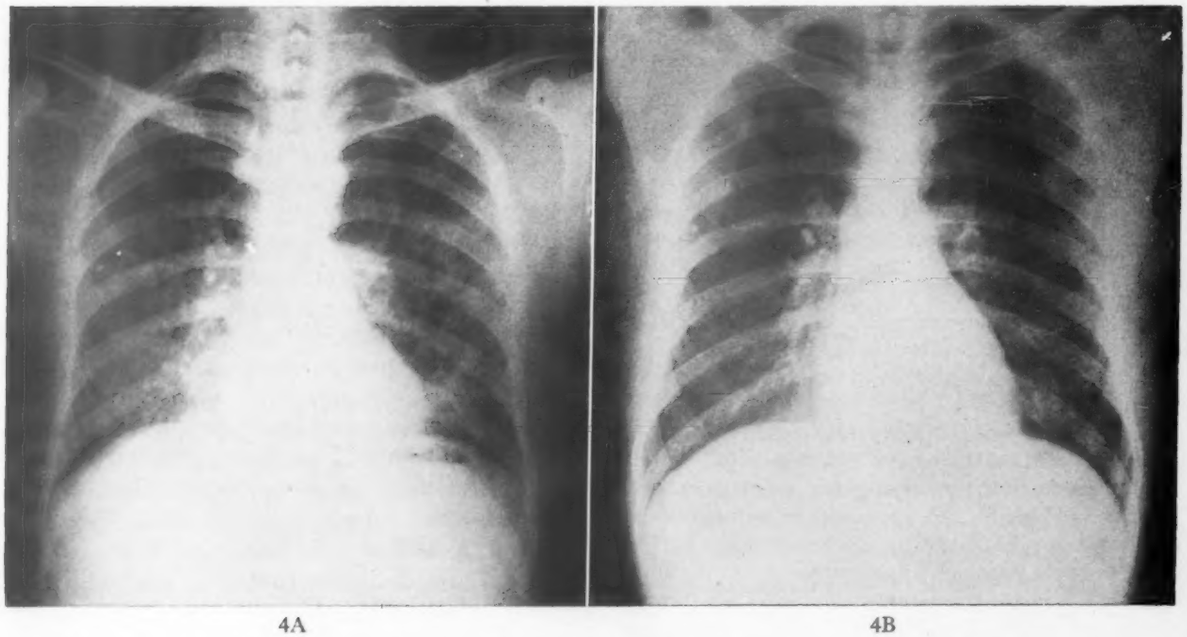
FIG. 2. Case II. A, chest roentgenogram taken on admission. B, follow-up examination three days later.

Urinalysis showed a trace of albumin and 1 plus sugar. The hemoglobin was 13.5 gm. per cent, the white blood cell count was 5,000 cells per cu. mm., with a normal differential. On admission, radiologic examination of the chest (Fig. 2A) revealed diffuse, patchy densities in both lung fields, radiating from the hilar areas. The size of the heart was difficult to evaluate since this roentgenogram was taken at the bedside in the anteroposterior projection. The impression was bronchopneumonia and the patient was treated with penicillin. Examination of the lungs six hours after admission showed clearing of rales with only scattered wheezes audible. The following day physical examination was within normal limits and a posteroanterior roentgenogram of the chest taken on the fourth hospital day (Fig. 2B) showed only promi-



3A 3B

FIG. 3. Case III. A, chest roentgenogram taken on admission. B, follow-up examination thirty hours later.



4A 4B

FIG. 4. Case IV. A, chest roentgenogram taken on admission. B, follow-up examination three days later.

nence of the right bronchovascular markings; urinalysis on the second day was normal.

CASE III. N. N., a twenty-six year old white heroin addict, on November 18, 1957 took an intravenous injection of heroin, immediately realized it was an overdose, and lost consciousness thereafter. Emergency treatment with caffein sodium benzoate was given on the way to the hospital. On admission the patient was raising large quantities of frothy, blood-tinged sputum. He appeared dyspneic, orthopneic, and cyanotic with a respiratory rate of 28 per minute; temperature, 100.4°F., pulse rate, 108 per minute; and blood pressure, 120/80 mm. Hg. The pupils were miotic and reacted sluggishly to light. The neck veins were distended and filled from below.

Examination of the chest revealed bubbling rales throughout both lung fields. The remainder of the physical examination revealed no abnormalities other than several venipuncture scars.

Urinalysis showed a trace of sugar and a trace of acetone. The hemoglobin was 17 gm. per cent; white blood count, 13,050 cells per cu. mm.; with a normal differential. An electrocardiogram revealed sinus tachycardia with right deviation of the electrical axis. On admission a posteroanterior roentgenogram of the chest (Fig. 3A) showed diffuse butterfly-shaped blotchy densities throughout both lung fields, leaving only a narrow clear zone in the periphery. The heart shadow was normal.

The patient received penicillin and tetracycline as well as 1.2 mg. of lanatoside C intravenously over the next four hours. Within six hours after admission, physical examination of the lungs was within normal limits and the patient felt completely well. A repeat x-ray film of the chest (Fig. 3B) taken thirty hours after admission showed some prominence of the bronchovascular markings, but was otherwise within normal limits.

Case IV. C. G., a thirty-two year old Puerto Rican man, entered Bellevue Hospital on November 1, 1957 because of cough of twelve hours' duration. The patient was a "main-line" heroin addict, who prior to admission followed overindulgence in alcohol with his customary injection of heroin and collapsed shortly thereafter. He regained consciousness about one hour later and noted a hacking cough productive of blood-tinged sputum. On admission to the hospital the patient seemed dyspneic, orthopneic, cyanotic and apprehensive. His vital signs were: respiratory rate, 24 per minute; temperature, 100°F; pulse rate, 110 per minute; blood pressure, 110/50 mm. Hg. Fine rales were scattered over both lung fields. There were multiple "main-line" scars on both upper extremities; otherwise physical examination was within normal limits.

Urinalysis was normal; hemoglobin, 15.5 gm. per cent; white blood cell count, 15,600 cells per cu. mm. with 90 per cent polymorphonuclear granulocytes on differential smear. A posteroanterior roentgenographic examination of the chest on admission (Fig. 4A) showed bilateral hilar congestion with diffuse mottled densities throughout the inner two-thirds of both lung fields. The heart was normal in size and appearance. Improvement was rapid; within twenty-four hours the physical examination was entirely within normal limits and the white blood cell count was 5,500 cells per cu. mm. A repeat roentgenographic examination of the chest (Fig. 4B) on the third hospital day revealed no abnormal findings.

COMMENTS

Heroin poisoning undoubtedly led to the clinical picture described in these four cases. Overdose effects may have been potentiated by the synergistic action of alcohol in one patient and by prolonged abstinence from the drug with loss of tolerance in another. A period of coma ranging from "a few minutes" to "one hour in length" followed the self-injection of heroin. Cyanosis, dyspnea, orthopnea and apprehension were apparent in all patients upon regaining consciousness. There was a cough productive of white mucoid or blood-tinged sputum. When seen at this hospital the respiratory rate was rapid in all patients, although severe respiratory depression was described prior to admission here. Although tachycardia was present in every case, the blood pressure remained within normal limits. A mild febrile response was noted in two patients. Distention of the neck veins, with filling from below, was apparent in one case whereas hepatomegaly, gallop rhythm and ankle edema were never observed. Auscultation of the lungs revealed the presence of wide-spread rales, variable in distribution. Glycosuria was present in two patients, a not unusual finding in morphine poisoning. The radiological manifestations were compatible with the diagnosis of pulmonary edema and had resolved when a follow-up film was taken sixteen to eighty hours later. As can be seen from this description, the only distinguishing features of a patient with this form of pulmonary edema are a history of opium alkaloid administration or the finding of an obtunded sensorium, miotic pupils, and/or multiple venipuncture scars on physical examination.

In the absence of the usual predisposing causes of pulmonary edema, the diagnosis was not always considered in these patients. Aspiration pneumonia, bronchopneumonia and pulmonary infarction were included among the various possibilities. Although the initial picture was suggestive of pneumonia, the rapid improvement within hours of admission as evidenced by the clearing of dyspnea, cyanosis and rales would favor a readily reversible transudation of edema fluid. This impression was confirmed by rapid radiologic resolution.

Pulmonary infarction was considered in view of the possible intravenous injection of heroin particles or of fibers from the cotton used by addicts as a filter. However, the lack of pain, physical findings or characteristic roentgenologic evidence make pulmonary embolism and infarction unlikely. Since the outcome was not fatal in any of the patients the diagnosis of pulmonary edema was not confirmed at autopsy. However,

congestion or frank edema of the lungs has been repeatedly reported as a postmortem finding in acute poisoning by opium alkaloids [*5,7,9,10,12*]. As a matter of fact, it is a consistent autopsy finding in acute heroin poisoning in the experience of the Medical Examiner's Office of New York City [*13*].

The multiple clinical and experimental conditions which precipitate pulmonary edema resolve into four final physiologic pathways which govern fluid dynamics at the capillary level: (1) capillary pressure; (2) capillary permeability; (3) plasma osmotic pressure; and (4) interstitial fluid pressure. The relative importance of these factors in pulmonary edema due to acute opium overdosage has not been studied experimentally and any attempt to explain the underlying mechanism is based on indirect evidence. The following known toxic effects of heroin * may act on these final pathways.

Respiratory Depression. Acute opium poisoning is associated with diminished ventilation and decreased arterial oxygen saturation [*11*]. The ensuing hypoxia may favor pulmonary edema by acting on pulmonary capillary permeability, pulmonary capillary pressure, or both. It has been suggested that altered capillary permeability is the principal determinant of pulmonary edema due to hypoxia [*14*]. However, the inaccessability of the pulmonary capillary bed to precise study by present technics precludes the final acceptance of this explanation [*4*], and different conclusions therefore have been reached by others [*1,16*], who favor pressure changes in the pulmonary circulation as being of greater importance in the genesis of hypoxic pulmonary edema.

The alternate mechanism of increased pulmonary capillary pressure in hypoxia can be derived from experiments in which heart failure appears during the course of severe hypoxia [*15–17*]. With heart failure, the increased left atrial pressure will be transmitted to the pulmonary capillaries through the valveless pulmonary veins of man. It is of interest in this respect that the administration of large doses of morphine in the cat results in an increase in pulmonary vein pressure [*1*]. Furthermore, it is possible that the pulmonary artery hypertension known to occur in anoxia [*19*] might be associated with hypertension in the pulmonary capillaries [*20*], thus promoting the development of pulmonary edema.

Central Nervous System Effects. Many disorders involving the central nervous system and characterized by increased intracranial pressure can lead to pulmonary edema [*1*]. Morphine, administered in therapeutic doses, is known to almost double the intracranial pressure [*21*] and has also been postulated to have a direct effect on the vasomotor center in the brain [*22*]. While the "neurohemodynamics" of pulmonary edema are by no means clear, it has been suggested that stimulation of the central nervous system, through peripheral vasoconstriction, results in an increase in pulmonary blood volume, and in a rise in pulmonary vascular pressure [*23*]. Since both of these changes favor transudation of fluid into the alveolar space an effect upon the central nervous system represents another possible mechanism of opium-induced pulmonary edema.

Histamine Liberation. The opium alkaloids lead to the liberation of histamine in the tissues of the rat [*24,25*], a substance which has been used in producing pulmonary edema experimentally, but is not known to have this effect in man. Since heroin poisoning is not associated with other clinical signs of histamine liberation, this constitutes a remote possibility in the genesis of pulmonary edema.

A Direct Action of Heroin on the Pulmonary Capillaries or on the Myocardium. There is no experimental evidence available to support such an effect, but conceivably a toxic action of heroin on the pulmonary capillary permeability or myocardium could result in pulmonary edema without invoking hypoxia or mediated mechanisms on the central nervous system.

Only limited therapeutic suggestions can be derived from these case reports. Until experimental evidence clarifies the pathophysiology of this form of pulmonary edema, treatment will remain empirical. Certainly, the use of N-allylnormorphine to counteract respiratory depression and oxygen to alleviate hypoxia seem important therapeutic measures and, judging from these four cases, would be sufficient. Should further experimental studies reveal heart failure to be of importance in this condition, the use of digitalis

* Most studies of this problem have been carried out with morphine. Heroin (diacetylmorphine) loses both acetyl groups and is hydrolyzed to morphine almost completely in the body, thereafter being metabolized and excreted like the sister alkaloid [*11*]. Experimental data obtained with morphine are therefore in all likelihood applicable to this discussion of heroin pulmonary edema.

and the usual measures to decrease venous return to the heart might be considered.

SUMMARY

1. Four cases of pulmonary edema secondary to acute heroin poisoning are reported. The characteristic features of this rare cause of pulmonary edema are mentioned.

2. The pathogenesis of heroin-induced pulmonary edema is not clear. It is a manifestation of acute opium alkaloid toxicity.

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(brand of norethynodrel with ethynylestradiol 3-methyl ether)

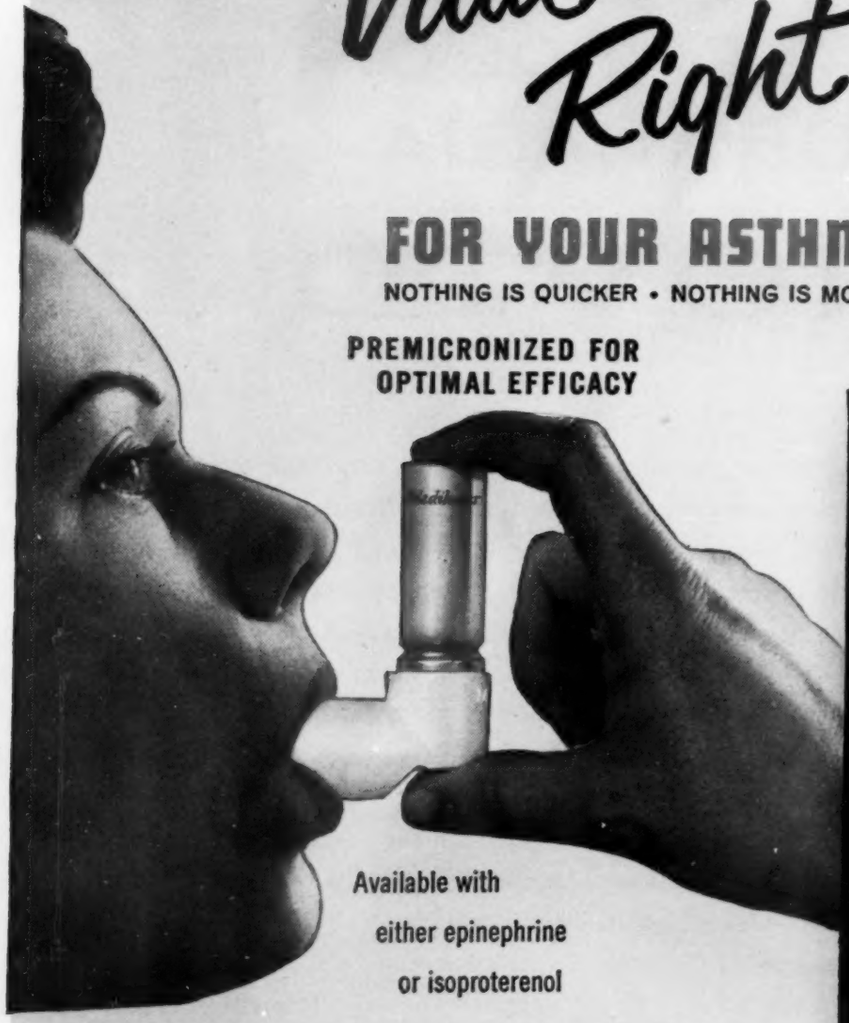
SEARLE | *Research in the Service of Medicine*

22½% More Vital Capacity Right Now

FOR YOUR ASTHMATICS

NOTHING IS QUICKER • NOTHING IS MORE EFFECTIVE

PREMICRONIZED FOR OPTIMAL EFFICACY



Available with either epinephrine or isoproterenol

Medihaler-EPI®

Epinephrine bitartrate, 7.0 mg. per cc., suspended in inert, nontoxic aerosol vehicle. Contains no alcohol. Each measured dose contains 0.15 mg. epinephrine.

Medihaler-ISO®

Isoproterenol sulfate, 2.0 mg. per cc., suspended in inert, nontoxic aerosol vehicle. Contains no alcohol. Each measured dose contains 0.06 mg. isoproterenol.

Riker Northridge, California

NOTABLY SAFE AND EFFECTIVE FOR CHILDREN, TOO.

MEDIHALER®

automatically measured-dose aerosol medications

Nonbreakable • Shatterproof • Spillproof • Leakproof

DIARRHEAS! DYSENTERIES!

For more than 20 years, Pectin has been the key therapeutic ingredient of pharmaceutical specialties used specifically for the treatment of diarrheas, dysenteries and other intestinal disorders. As early as 1933, the detoxicating effect of Pectin was attributed to its galacturonic acid content.

PECTIN IN THE NAME IS NOT ENOUGH

Adequate dosage is essential. It is the galacturonic acid intake (2 to 4 grams of Pectin N.F. per day) that determines the effectiveness. In many instances the use of relatively inert adsorbent fillers has limited the amount of the therapeutic detoxicant, PECTIN, in the formulation to an inadequate dosage.

Exchange Brand Pectin N.F.

Increases bulk and fluid retention of upper intestinal contents and imparts a smooth, gelatinous consistency • Lubricates the intestinal wall • Promotes normal peristalsis without mechanical irritation • Reduces intestinal pH • Inhibits growth of many putrefactive and otherwise undesirable microorganisms in the intestines without affecting normal flora • Promotes assimilation of essential nutrients • Helps to conjugate and eliminate toxins • Reduces toxic side effects of therapeutic agents.

Exchange Brand Citrus Pectin and Pectin derivatives widely used in therapeutic specialties include:

PECTIN N.F. • POLYGALACTURONIC ACID • PECTIN CELLULOSE COMPLEX • GALACTURONIC ACID • *These are available to the medical profession in specialties of leading pharmaceutical manufacturers.*

Sunkist Growers

PRODUCTS SALES DEPARTMENT
PHARMACEUTICAL DIVISION
ONTARIO, CALIFORNIA

Oatmeal

Leads all other Whole-Grain Cereals in Quantity *and* Quality of Protein

The quality of protein is graded by several criteria of which the most frequently used are: total protein value* and protein efficiency ratio.** Independently published data[1] reveal that oatmeal excels on both counts:

	Total Protein Value	Protein Efficiency Ratio
Oatmeal	165	2.2
Whole wheat	140	1.5
Barley	87	1.8
Whole corn	84	1.2
Rice	57	1.9

Quantitatively, the customary oatmeal-with-milk dish (¾ cup cooked oatmeal and 4 fluid ounces milk) provides 9 grams of protein, more than that provided by any other whole-grain cereal with milk.

In addition to its qualitative and quantitative superiority in protein, oatmeal-and-milk supplies significant amounts of B vitamins and minerals to complement the excellent nutritive balance between protein, fat and carbohydrate.

Just as important to the physician who prescribes a cereal food is oatmeal's delicious nut-like flavor, its ease of digestion, and the ready availability of its contained nutrients. Whether it be for an infant's first solid feeding, for the geriatric patient, or in many other situations, oatmeal makes a real contribution to the day's nutritional needs.

Nutrition studies of The Quaker Oats Company Research Laboratories assure that Quaker Oats and Mother's Oats, the two brands of oatmeal marketed by The Quaker Oats Company, are practically identical with natural whole-grain oats in composition and in nutritive values. Both brands are available in the Quick (cooks in one minute) and the Old-Fashioned varieties which are of equal nutrient value.

*Total protein value is determined by multiplying biologic value x digestibility x per cent nitrogen.

**Protein efficiency ratio is the efficiency with which an experimental animal utilizes each gram protein consumed.

1. Rand, N.T., and Collins, V.K.: Food Technology *12*:585 (Nov.) 1958.

The Quaker Oats Company
CHICAGO

because it "most closely approaches the 'ideal anticoagulant,'"[1] COUMADIN has become widely accepted as the anticoagulant of choice[1-3]

"most of the drawbacks of Dicumarol have been overcome [with COUMADIN] ... It is my firm belief that in time it will replace Dicumarol on the basis of its performance over a wide variety of conditions..."[2]

COUMADIN®

ORAL
I.V. or I.M.

SODIUM

IN MYOCARDIAL INFARCTION AND OTHER THROMBOEMBOLIC DISORDERS

After more than five years' clinical experience, it has been concluded: "In my opinion, Warfarin sodium [COUMADIN] is the best anticoagulant available today."[3]

COUMADIN CONSISTENTLY PROVIDES

rapid and sustained effect with low dosage • high predictability • ease of control for long periods • low incidence of "escape" • equal effectiveness by oral or parenteral routes • reduced need for frequent prothrombin time determinations after initial dosage adjustment • ready reversibility with vitamin K_1

Complete Information and Reprints on Request

Endo®
ENDO LABORATORIES
Richmond Hill 18, New York

TABLETS
For oral administration—2 mg., lavender, scored; 5 mg., peach, scored; 10 mg., white, scored; 25 mg., red, scored.

INJECTION
For parenteral administration—Single Injection Units, consisting of one vial, 75 mg., and one 3-cc. ampul Water for Injection.

AVERAGE DOSE
Initial, 50 mg. *Maintenance*, 5-10 mg. daily, as indicated by prothrombin time determinations.

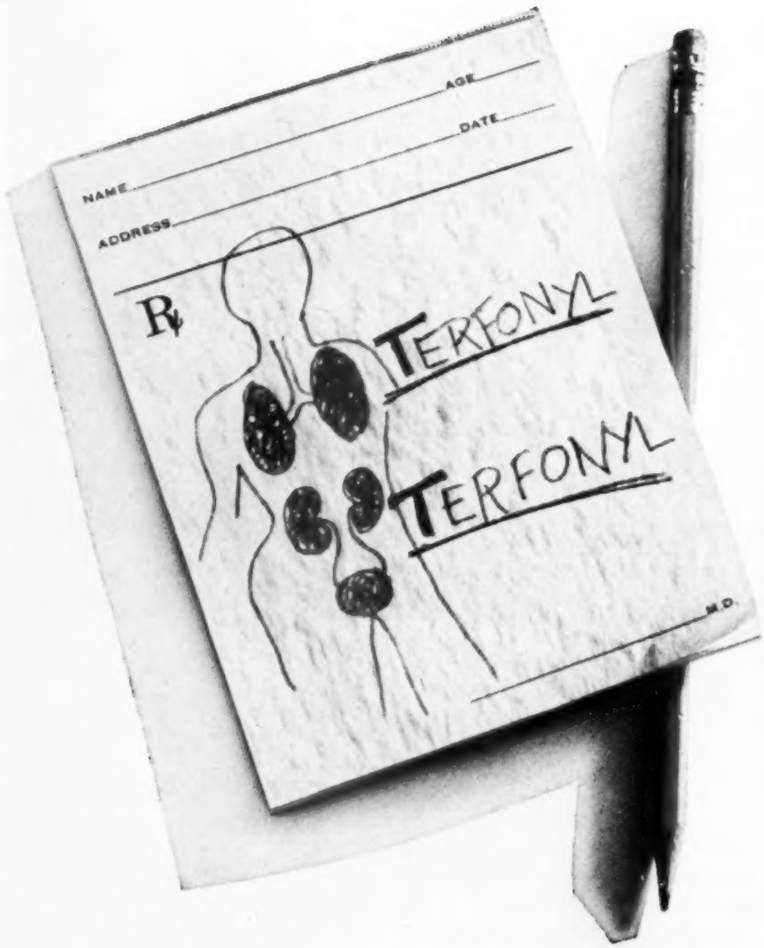
COUMADIN (warfarin) Sodium—manufactured under license from the Wisconsin Alumni Research Foundation—*developed for clinical use by Endo.*

References: 1. Baer, S., et al.: J.A.M.A. 167:704, 1958. 2. Link, K. P.: Circulation 19:97, 1959. 3. Meyer, O. O.: Postgrad. Med. 24:110, 1958.

think of Terfonyl first

Squibb Triple Sulfas Trisulfapyrimidines

THE TRIPLE SULFONAMIDE OF CHOICE IN COMMON URINARY, RESPIRATORY, AND OTHER BACTERIAL INFECTIONS



for effectiveness...
Prompt, high blood levels[1,2]
Excellent tissue diffusion[3]
Broad in scope—attacks both gram-positive and gram-negative pathogens[2,3]

for safety...
Highly soluble in urine—especially at critical pH levels[2]
Impressively low incidence of sensitization reactions[2,3]
Frequently avoids the problem of the development of resistant organisms

for economy...
Terfonyl is economical to prescribe for your patients

for palatability...
Good tasting, raspberry-flavored suspension appealing to all age groups—particularly the young and elderly patients

Supply:
Raspberry-flavored suspension, containing 0.167 Gm. each of sulfamethazine, sulfadiazine, and sulfamerazine per teaspoonful (5 cc.). Pint bottles.

Tablets, containing 0.167 Gm. each of sulfamethazine, sulfadiazine, and sulfamerazine per 0.5 Gm. tablet. Bottles of 100 and 1000.

Also available:
Pentid-Sulfas: Each tablet contains 200,000 units of crystalline penicillin G potassium and 0.167 Gm. each of sulfamethazine, sulfadiazine, and sulfamerazine. Bottles of 30, 100 and 500.

References:
1. Campbell, M. F.: Principles of Urology, Philadelphia, W. B. Saunders Company, 1957, p. 232. 2. Sophian, L. H.; Piper, D. L., and Schneller, G. H.: The Sulfapyrimidines, New York, A. Colish, 1952, pp. 70-76. 3. Lehr, D.: New York J. Med. 50:1361 (June 1) 1950.

SQUIBB

Squibb Quality / the Priceless Ingredient

"TERFONYL®" AND "PENTIDS®" ARE SQUIBB TRADEMARKS

FOR PROVEN MENOPAUSAL BENEFITS
with extra relief from anxiety and tension

The vast majority of menopausal women, *especially on the first visit,* are nervous, apprehensive, and tense. PMB-200 or PMB-400 gives your patient the advantage of *extra* relief from anxiety and tension, particularly when the patient is "high strung," under prolonged emotional stress, or when psychogenic manifestations are acute.

Proven menopausal benefits are confirmed by the wide clinical acceptance of "Premarin," specifically for the relief of hot flushes and other symptoms of estrogen deficiency, together with the well established tranquilizing efficacy of meprobamate.

Two potencies to meet the needs of your patients:

PMB 200

"PREMARIN"® WITH MEPROBAMATE*

PMB-200—Each tablet contains conjugated estrogens equine ("Premarin") 0.4 mg., and 200 mg. of meprobamate. When greater tranquilization is necessary you can prescribe PMB-400 – Each tablet contains conjugated estrogens equine ("Premarin") 0.4 mg., and 400 mg. of meprobamate. Both potencies are available in bottles of 60 and 500.

AYERST LABORATORIES
New York 16, N.Y., Montreal, Canada

*MEPROBAMATE, LICENSED UNDER U. S. PAT. NO. 2,724,720.

NEW THERAPY...TO DEFEAT THE MIGRAINE PARADOX*

'MIGRAL'®

TRADEMARK

• relieves headache
• dispels visual disturbances
and • overcomes nausea and vomiting

*The paradox of migraine – increased nausea due to ergotamine administration – may now be successfully combated with 'Migral'. The recognized benefits of ergotamine and caffeine in 'Migral' are favorably enhanced by the addition of cyclizine hydrochloride, a specific to overcome nausea.

Dosage: 2 to 3 tablets at first warning of an attack, then 1 or 2 tablets every half hour; not more than 6 tablets should be taken for any single attack.

Supplied: 'Migral' tablets, containing ergotamine tartrate 1 mg., 'Marezine'® brand Cyclizine Hydrochloride 25 mg., and caffeine 50 mg.

BURROUGHS WELLCOME & CO. (U.S.A.) INC., Tuckahoe, New York

Paradox of the scalp-scratcher: Why will he scratch here?

Sad but true: the one man who can help him is the last man he'll ask—you. (Dandruff a *disease?*) Result: he goes right on scratching and hoping . . . and looking and scratching. You can end his search and gain his lasting appreciation, with a little medical advice—and a prescription for Selsun. Abbott

SELSUN®
(Selenium Sulfide, Abbott)

an ethical answer to a medical problem

anemia of pregnancy, **another indication**

for filmtab® **Iberol®**: potent antianemia therapy

plus the complete B-complex

JUST 2 IBEROL FILMTABS A DAY SUPPLY

The Right Amount of Iron

Ferrous Sulfate, U.S.P.	1.05 Gm.
(Elemental Iron—210 mg.)	

Plus the Complete B Complex

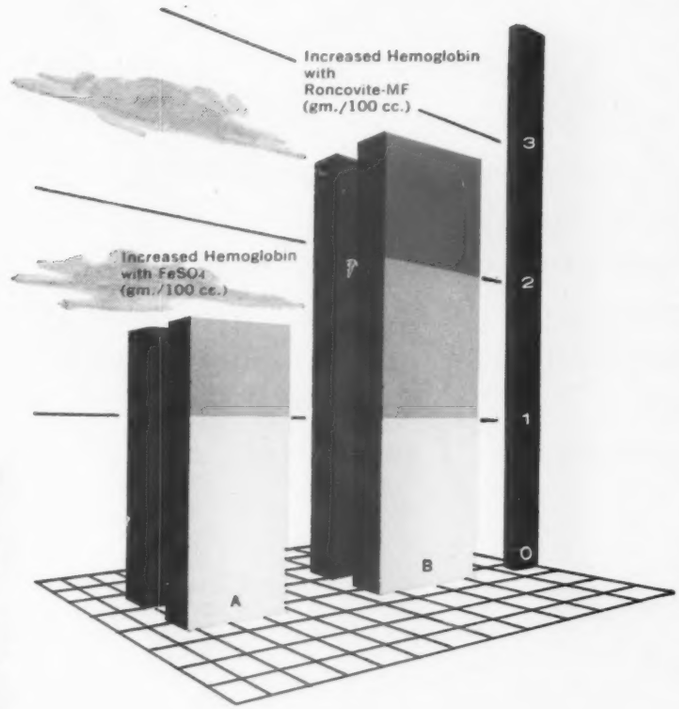
Vitamin B_{12} with Intrinsic Factor Concentrate	1 U.S.P. Unit (Oral)
Folic Acid	2 mg.
Liver Fraction 2, N.F.	200 mg.
Thiamine Mononitrate	6 mg.
Riboflavin	6 mg.
Nicotinamide	30 mg.
Pyridoxine Hydrochloride	3 mg.
Calcium Pantothenate	6 mg.

Plus Vitamin C

Ascorbic Acid	150 mg.

®FILMTAB—FILM-SEALED TABLETS, ABBOTT; U.S. PAT. NO. 2,881,085. © 1959, ABBOTT LABORATORIES 907020

Abbott


LEGEND:
(A) Patients receiving ferrous sulfate 200 mg. q.i.d. showed average increase in hemoglobin of 1.5 gm.
(B) Patients receiving Roncovite-MF (15 mg. cobalt chloride and 100 mg. ferrous sulfate) showed average increase in hemoglobin of 2.7 gm.
Increased Hemoglobin with Roncovite-MF (gm./100 cc.)
Increased Hemoglobin with FeSO4 (gm./100 cc.)
3
2
1
0
A
B

Improved iron utilization in anemia

RONCOVITE®-mf

Each tablet contains: Cobalt chloride (Cobalt as Co....3.7 mg.)...15 mg. Ferrous sulfate, exsiccated...100 mg.

Improves iron utilization by enhancing the formation of erythropoietin, the erythropoietic hormone

Recent research[1,2] again emphasizes the role of cobalt as the only clinically proved agent which enhances erythropoietin formation.

The acceleration of erythropoiesis with Roncovite therapy provides optimal utilization of iron.

Acting through this physiologic mechanism, Roncovite (cobalt-iron) therapy results in an increased production of red cells and hemoglobin—a better blood picture—a faster, more complete response than iron alone in the common hypochromic anemias—menstrual anemia—anemia of pregnancy—nutritional anemia of infancy—and in anemia due to chronic infection or inflammation.[3,4,5,6,7,8]

(1) Goldwasser, E.; Jacobson, L. O.; Fried, W., and Plzak, L. F.: Blood *13*:55 (Jan.) 1958. (2) Gurney, C. W.; Jacobson, L. O., and Goldwasser, E.: Ann. Int. Med. *49*:363 (Aug.) 1958. (3) Korst, D. R.; Bishop, R. C., and Bethell, F. H.: J. Lab. & Clin. Med. *52*:364 (Sept.) 1958. (4) Ausman, D. C.: Journal-Lancet *76*:290 (Oct.) 1956. (5) Holly, R. G.: Obst. & Gynec. *9*:299 (Mar.) 1957. (6) Holly, R. G.: Clin. Obst. & Gynec. *1*:15 (Mar.) 1958. (7) Diamond, E. F.; Gonzales, F., and Pisani, A.: Illinois M. J. *113*:154 (April) 1958. (8) Hill, J. M.; La Jous, J., and Sebastian, F. J.: Texas J. Med. *51*:686 (Oct.) 1955.

LLOYD BROTHERS, INC. • **CINCINNATI 3, OHIO**

To the relief of musculoskeletal pain, new MEDAPRIN* adds restoration of function

Analgesics offer temporary relief of musculoskeletal pain, but they merely *mask* pain rather than getting at its *cause*. New Medaprin, in addition to bringing about prompt subjective improvement, promotes the *restoration of normal function* by suppressing the inflammation that *causes* the pain.

Medaprin, Upjohn's new analgesic-steroid combination, contains aspirin plus Medrol,** the corticosteroid with *the best therapeutic ratio in the steroid field.*† Instead of suffering recurrent discomfort because of the "wearing off" of analgesics, the patient on Medaprin experiences a smooth, *extended* relief and more normal mobility.

Indications: Medaprin is indicated in mild-to-moderate rheumatic and musculoskeletal conditions, including rheumatoid arthritis, deltoid bursitis, low back pain, neuralgia, synovitis, fibromyositis, osteoarthritis, low back sprain, traumatic wrist, sciatica, and "tennis elbow."

Dosage: The recommended dosage is 1 tablet q.i.d. The usual cautions and contraindications of corticotherapy should be observed.

Supplied: In bottles of 100 and 500.

Formula: Each Medaprin tablet contains

- 300 mg. acetylsalicylic acid, for prompt relief of pain
- 1 mg. Medrol, to suppress the causative inflammation
- 200 mg. calcium carbonate, as buffer

*TRADEMARK **TRADEMARK, REG. U. S. PAT. OFF.—METHYLPREDNISOLONE, UPJOHN
†RATIO OF DESIRED EFFECTS TO UNDESIRED EFFECTS

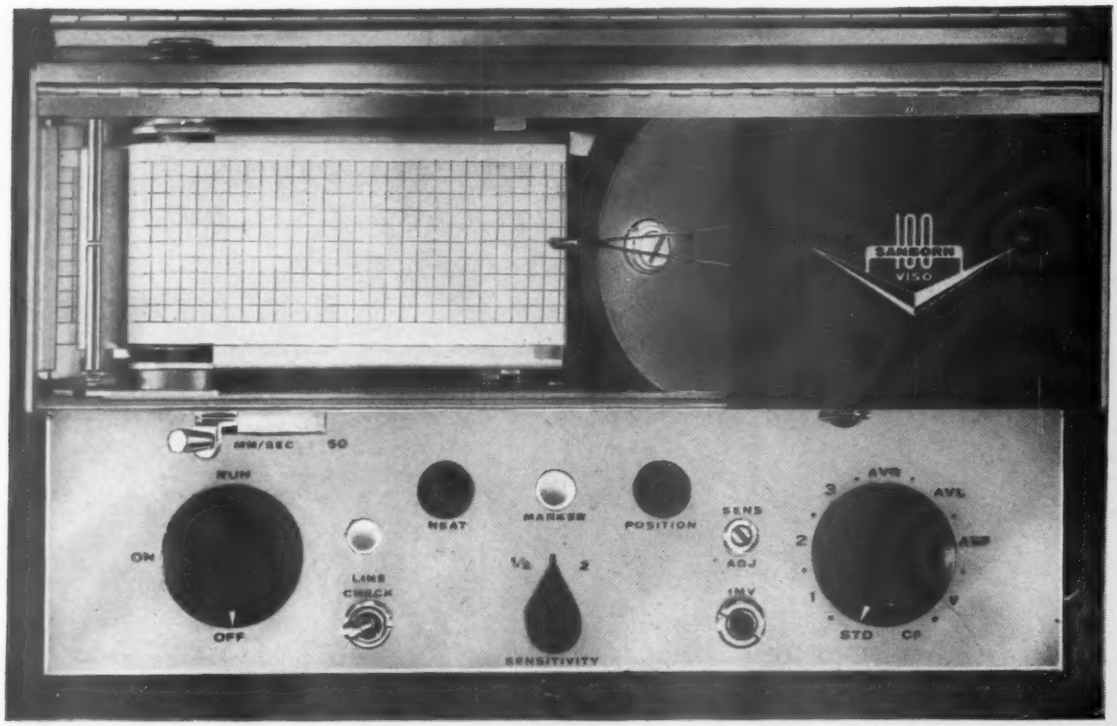
The Upjohn Company, Kalamazoo, Michigan

Upjohn

The new Sanborn 100 Viso electrocardiograph: two speeds . . . 25 or 50 mm/sec . . . clearly defined, permanent traces on 6cm charts . . . normal, ½, or 2-times recording sensitivity . . . two additional inputs for recording other phenomena, plus outlet for connecting monitoring oscilloscope . . . 15 transistors saving space, weight, and power . . . and the mobility of 29 pounds, complete . . . make this

FUNCTIONALLY AND FACTUALLY

the most modern electrocardiograph you can own



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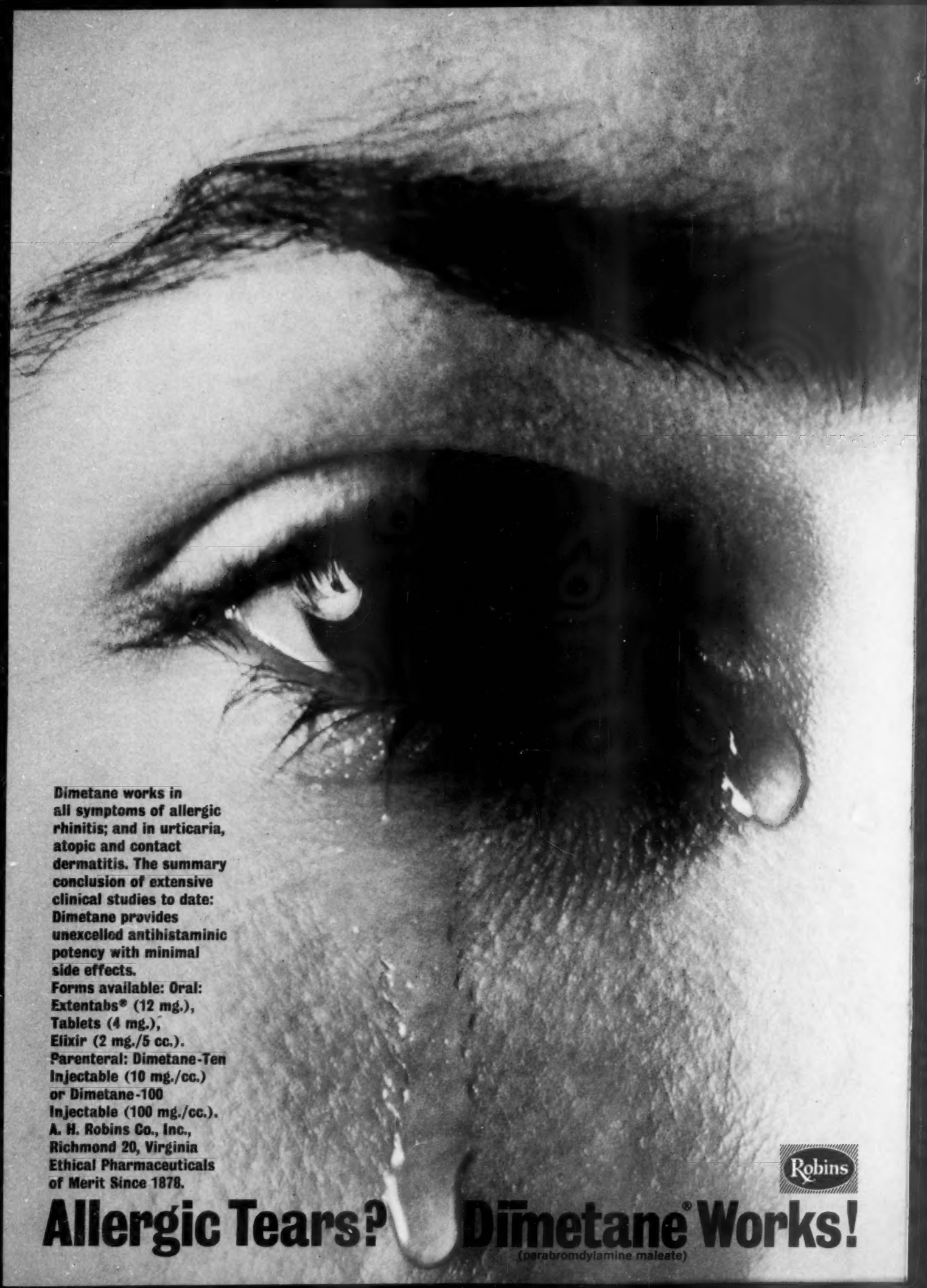


Dimetane works in all symptoms of allergic rhinitis; and in urticaria, atopic and contact dermatitis. The summary conclusion of extensive clinical studies to date: Dimetane provides unexcelled antihistaminic potency with minimal side effects.
Forms available: Oral: Extentabs® (12 mg.), Tablets (4 mg.), Elixir (2 mg./5 cc.). Parenteral: Dimetane-Ten Injectable (10 mg./cc.) or Dimetane-100 Injectable (100 mg./cc.).
A. H. Robins Co., Inc., Richmond 20, Virginia
Ethical Pharmaceuticals of Merit Since 1878.

Robins

Allergic Tears? Dimetane® Works!

(parabromdylamine maleate)

In peptic ulcer,

five aids to comprehensive management

with 1 *preparation*

Added to the therapeutic regimen, ALUDROX SA simplifies your comprehensive management of the peptic-ulcer patient. With ALUDROX SA you can relieve the patient's pain, reduce his acid secretion, inhibit gastric motility, calm his emotional distress, and promote healing of his ulcer.

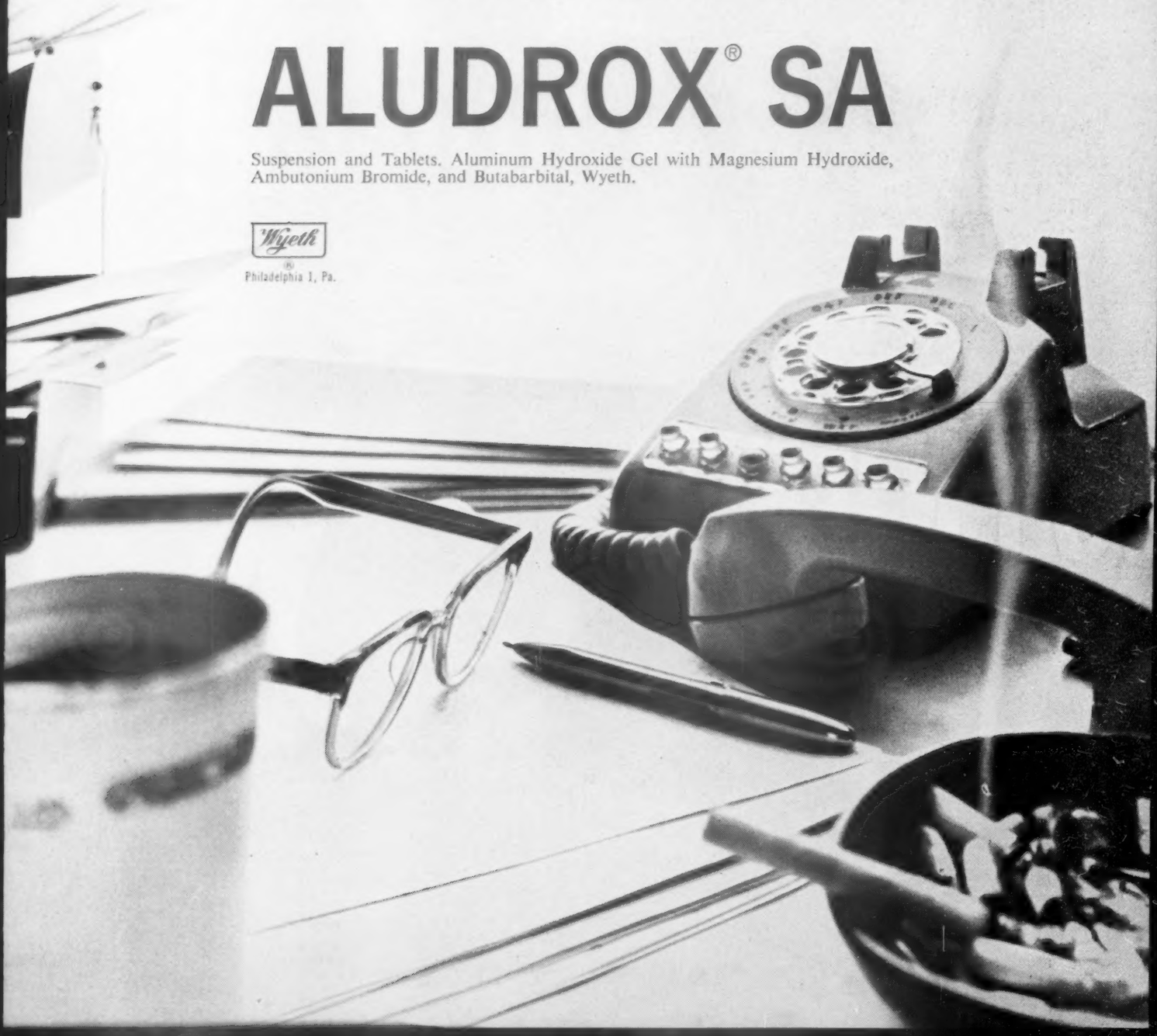
Ambutonium, an important new anticholinergic of demonstrated usefulness, is incorporated in ALUDROX SA to provide potent antisecretory and antimotility effects without significant side-reactions.

anticholinergic • antacid • sedative • anticonstipant • pepsin-inhibitor

ALUDROX® SA

Suspension and Tablets. Aluminum Hydroxide Gel with Magnesium Hydroxide, Ambutonium Bromide, and Butabarbital, Wyeth.

Wyeth®
Philadelphia 1, Pa.



the **"Stylish Stout"**
who'd like a
"Junior Miss"
figure

To help her slim down to "bathing beauty" size—for her health's sake as well as for her social need—

SYNDROX®

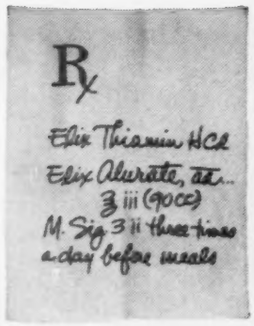
methamphetamine hydrochloride

- controls the desire for constant nibbling—suppresses appetite
- takes the mind off food by encouraging activity and a brighter viewpoint
- Dose for appetite suppression: ½ to 1 tab. or tsp. before meals.

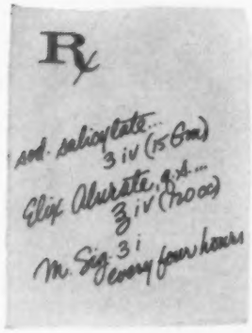
SYNDROX TABLETS (5 mg.)
ELIXIR (5 mg. per 5 cc.)

McNEIL McNEIL LABORATORIES, INC. PHILADELPHIA 32, PA.

Whenever your Rx requires a sedative component...



Rx FOR NERVOUS ANOREXIA



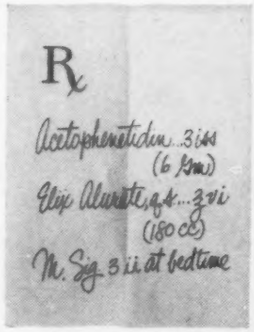
Rx FOR NEURITIC PAIN

widely compatible

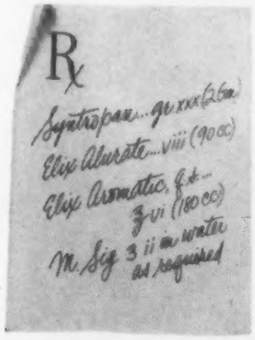
Alurate

sedative-hypnotic

Combines prompt action, broad margin of safety and virtual freedom from after-effects in a host of common conditions



Rx FOR INSOMNIA DUE TO PAIN



Rx FOR SPASTIC DYSMENORRHEA

To avoid identification or abuse of the barbiturate component, Alurate is available as Elixir Alurate (cherry-red) and Elixir Alurate Verdum (emerald-green).

ALURATE® brand of aprobarbital.
ROCHE®

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Classic therapy
for preventing and healing
diaper rash
excoriation, chafing, irritation

DESITIN®
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. . . enduring in its efficacy
. . . pleasing in its simplicity
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SAMPLES on request DESITIN CHEMICAL COMPANY
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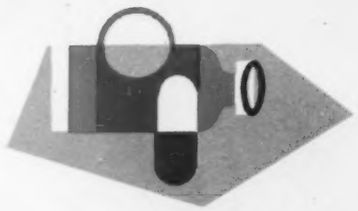
three essential steps help overweight patients eat to live, not live to eat

supervision by the physician

a balanced eating plan



supportive medication

Obedrin®

and the 60-10-70 Basic Plan

provide an effective weight control regimen

Frequently a patient loses weight while on a special diet, then soon gains it back again. Obedrin is a valuable aid to this type of patient. It curbs unhealthy food craving while the patient establishes correct eating habits. Thus he becomes able to *maintain* optimum weight.

Each capsule or tablet provides:

Semoxydrine® HCl (methamphetamine HCl), 5 mg., for its anorexigenic and mood-lifting effects

Pentobarbital, 20 mg., to guard against excitation

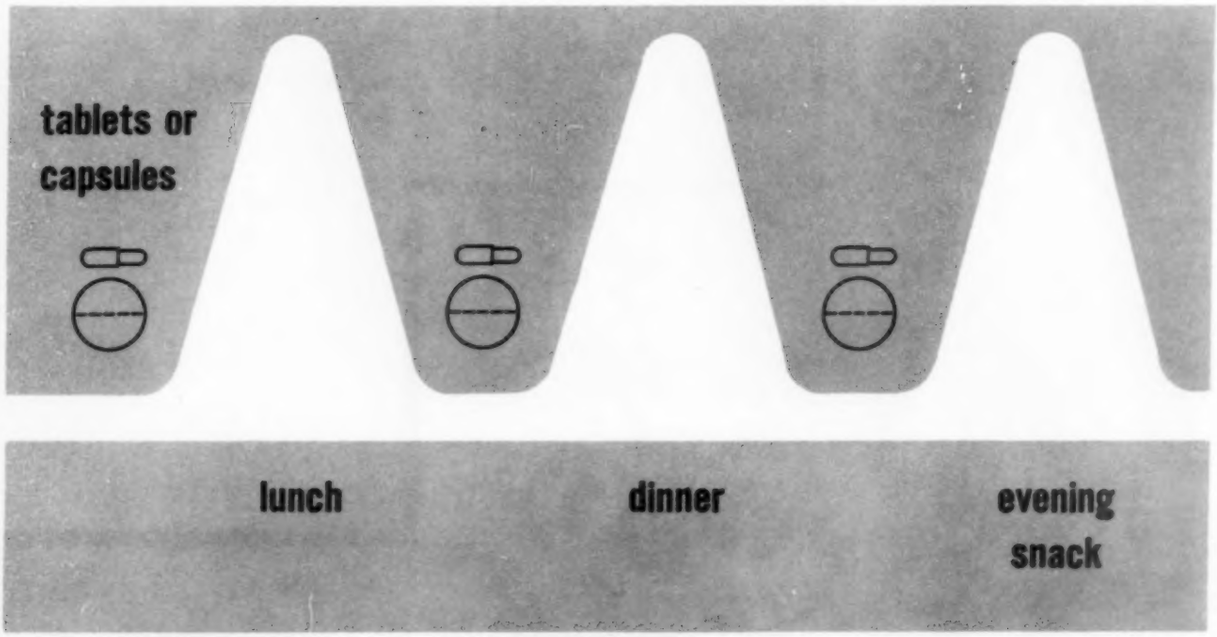
Thiamine Mononitrate, 0.5 mg., Riboflavin, 1 mg. and Nicotinic Acid (Niacin), 5 mg., to supplement the diet

Ascorbic Acid, 100 mg., to help mobilize tissue fluids

Bristol, Tennessee • New York • Kansas City • San Francisco THE S. E. MASSENGILL COMPANY

for dependable control of appetite

. . . a flexible dosage form



The Obedrin formula permits a flexible dosage schedule which depresses the appetite when it is most important to do so—at peak hunger periods. The physician can adjust the dosage to fit each patient's need.

Obedrin®

and the 60-10-70 Basic Plan

advantages of Obedrin

A dependable anorexigenic agent

A flexible dosage form

Minimal central nervous stimulation

Vitamins to supplement the diet

No hazards of impaction

Write for 60-10-70 menus, weight charts, and samples of Obedrin.

Used with the 60-10-70 Basic Plan, Obedrin offers an ideal weight-control regimen for the overweight patient.

Bristol, Tennessee • New York • Kansas City • San Francisco THE S. E. MASSENGILL COMPANY

the first *oral* antifungal antibiotic for ringworm

soon available

*T.M. 5-224

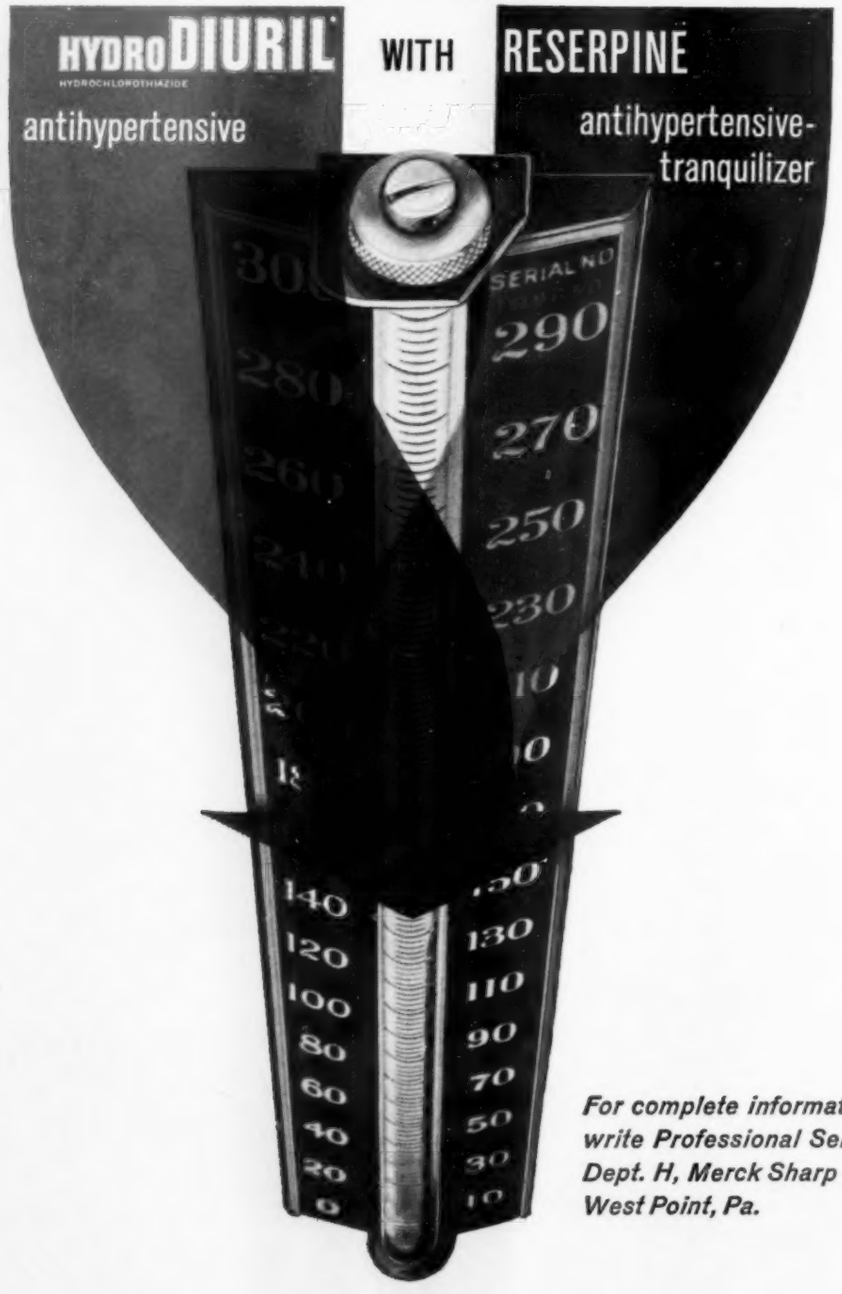
greater antihypertensive effect...fewer side effects

HYDROPRES*

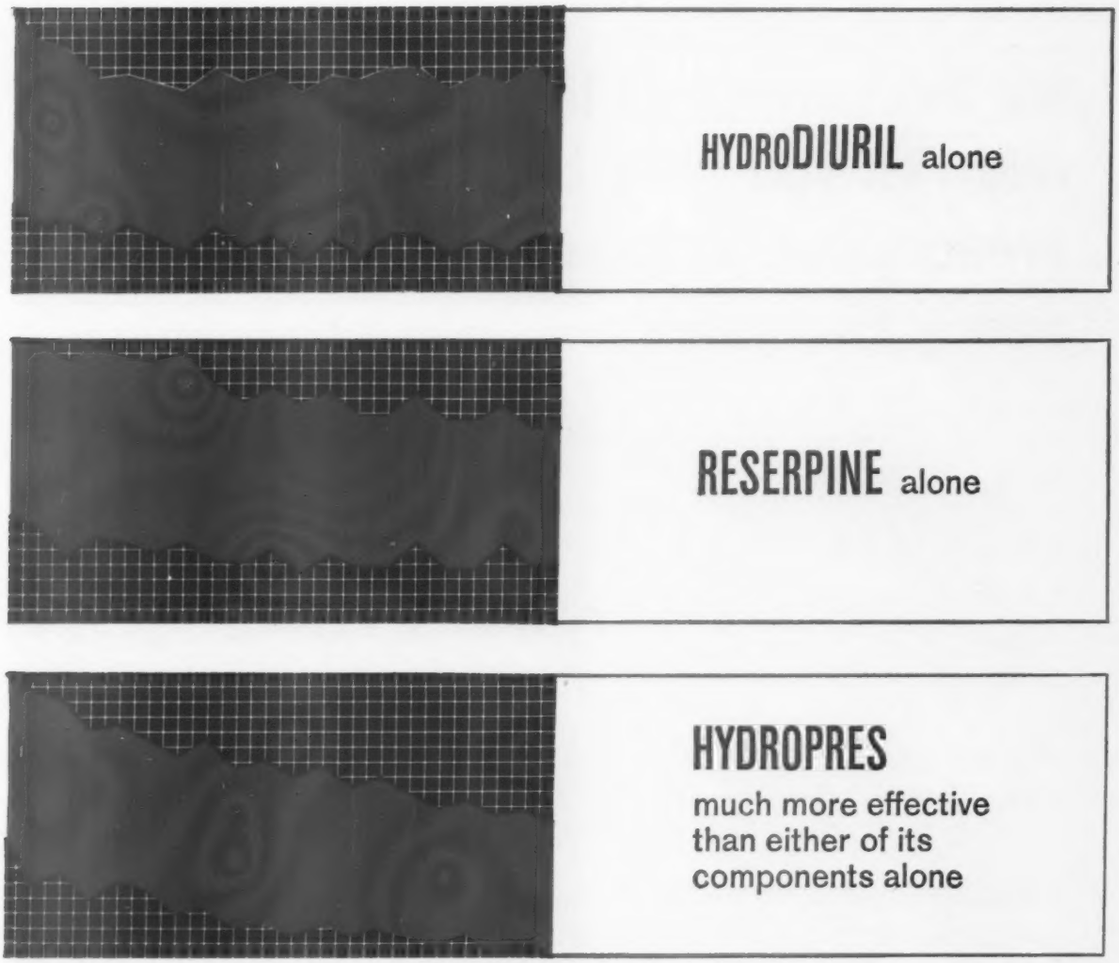
HYDRODIURIL® HYDROCHLOROTHIAZIDE antihypertensive

WITH

RESERPINE antihypertensive-tranquilizer



For complete information write Professional Services, Dept. H, Merck Sharp & Dohme, West Point, Pa.



- Effective by itself in a majority of patients. Provides smooth, more trouble-free management of hypertension.
- Since HYDRODIURIL and reserpine potentiate each other, the required dosage of each is lower when given together as HYDROPRES than when either is given alone.
- HYDROPRES provides the needed and valuable tranquilizing effect of reserpine. Lower dosage may reduce such side effects of reserpine as excessive sedation and depression.
- Arrest or reversal of organic changes of hypertension may occur.
- Headache, dizziness, palpitations and tachycardia are usually promptly relieved. Anginal pain may be reduced in incidence and severity.
- With HYDROPRES, dietary salt may be liberalized.
- Convenient, controlled dosage.

HYDROPRES-25

25 mg. HYDRODIURIL, 0.125 mg. reserpine.
One tablet one to four times a day.

HYDROPRES-50

50 mg. HYDRODIURIL, 0.125 mg. reserpine.
One tablet one or two times a day.

If the patient is receiving ganglion blocking drugs or hydralazine, their dosage must be cut in half when HYDROPRES is added.

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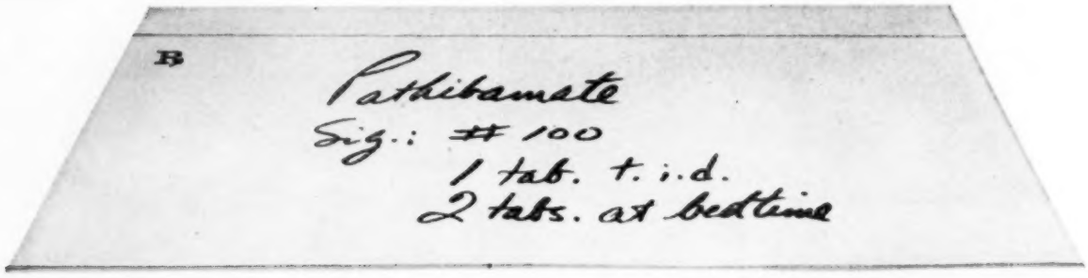

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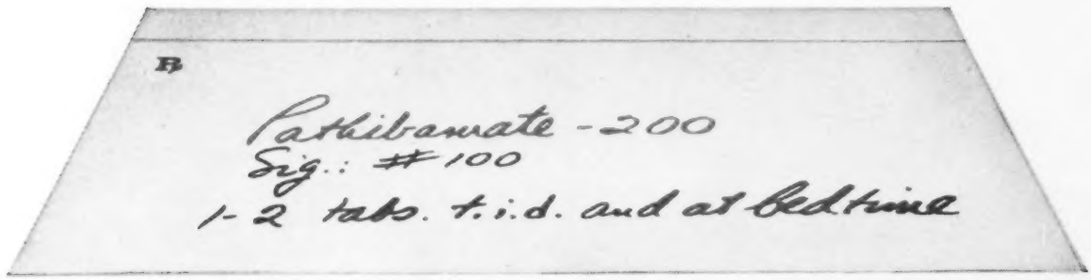

In the management of such gastrointestinal dysfunctions as duodenal or gastric ulcer, intestinal colic, spastic and irritable colon, ileitis, esophageal spasm, gastric hypermotility and anxiety neurosis with G. I. symptoms, nearly two years' experience has confirmed the clinical advantages derived from the combination of the two agents in PATHIBAMATE.

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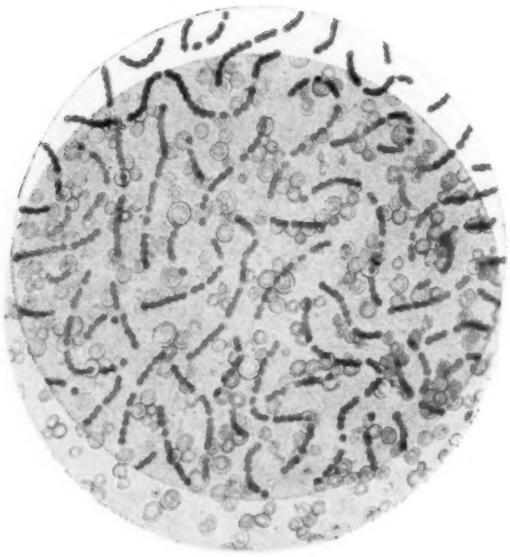

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Specific for skeletal muscle spasm
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The analgesic preferred in musculoskeletal pain

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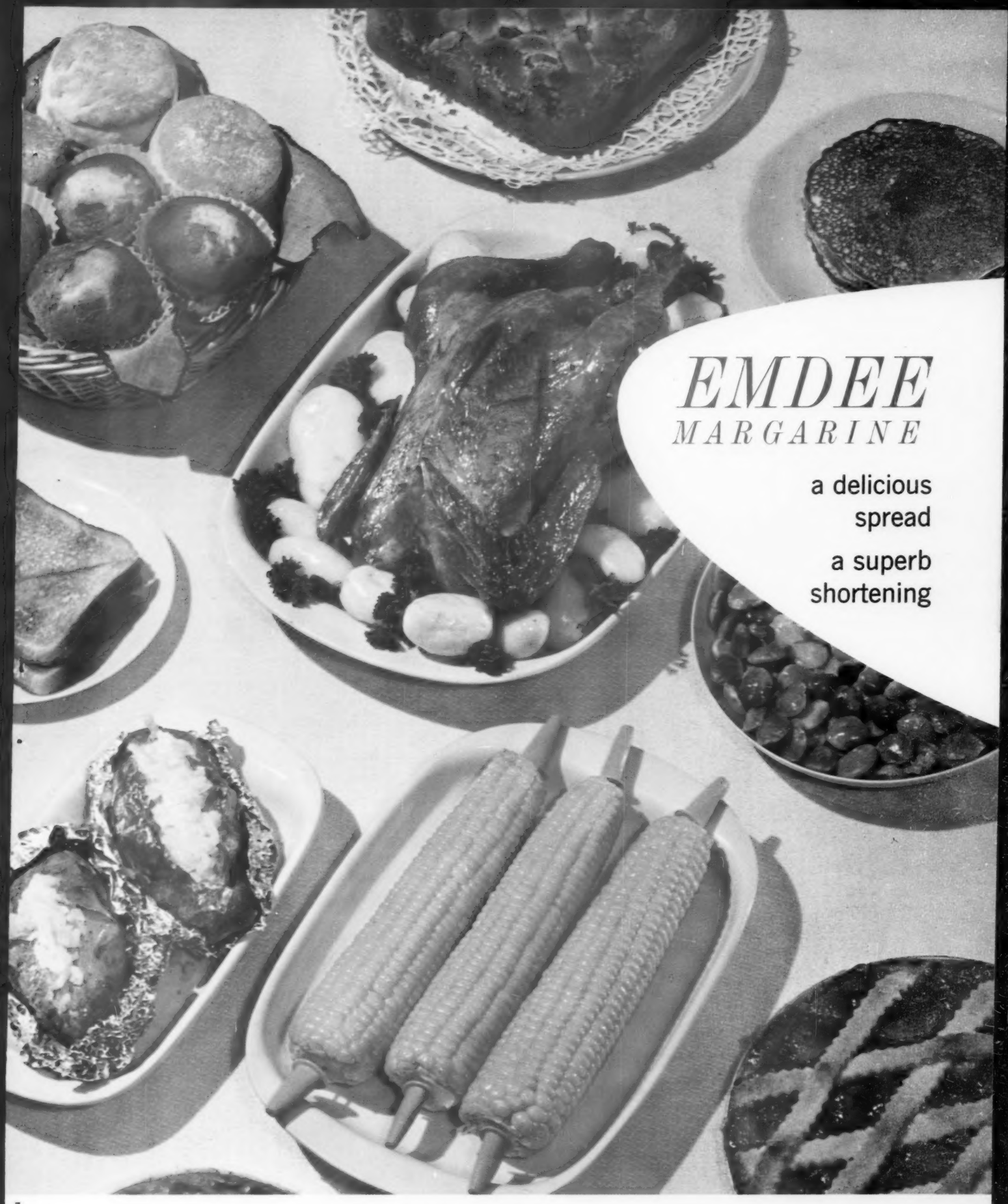

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References: 1. Terman, L. A.: Dietary management of hypercholesterolemia, Geriatrics *14*:111 (Feb.) 1959. 2. Boyer, P. A.; Lowe, J. T.; Gardier, R. W., and Ralston, J. D.: A new dietary management of hypercholesterolemia, J.A.M.A., in press. 3. Vail, Gladys E.: Cooking with fats high in polyunsaturated fatty acids, J. Am. Dietet. A. *35*:119 (Feb.) 1959.

Reprints of these articles on Emdee Margarine are available on request.

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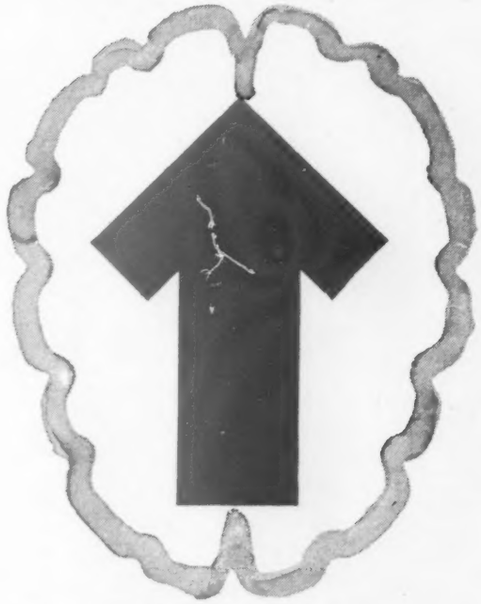
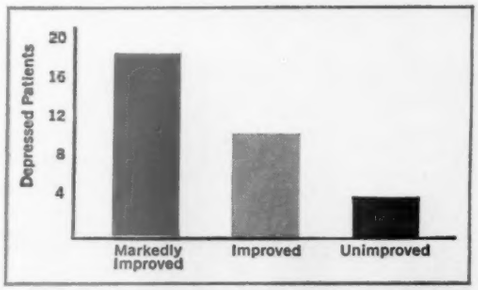


Revitalizes depressed patients—elevates mood, increases alertness and ability to maintain work and social adjustment.[1,2]



1. Agin, H. V.: In A Pharmacologic Approach to the Study of the Mind, Springfield, Ill., Charles C Thomas, In press.
2. Agin, H. V.: Conference on Amine Oxidase Inhibitors, New York Academy of Sciences, Nov. 20-22, 1958.

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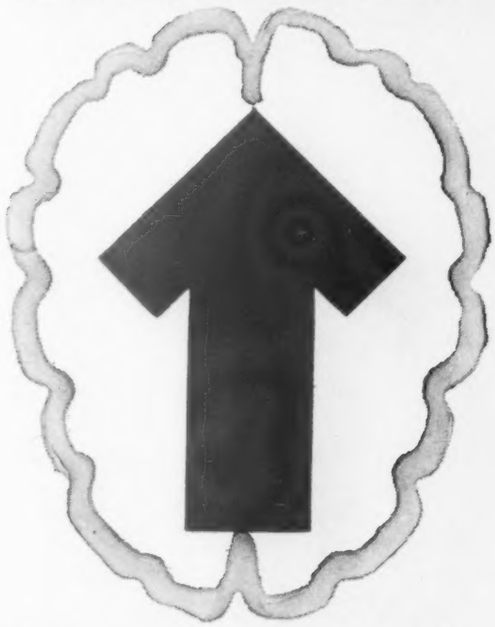
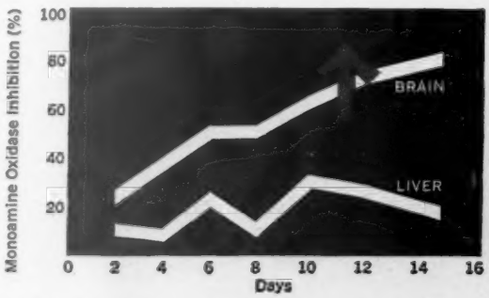


Brightens mood, dispels apathy, melancholy, social withdrawal through selective suppression of monoamine oxidase (MAO) of brain at doses which have little or no effect on liver.



Horita, A.: Report, Mar. 17, 1959

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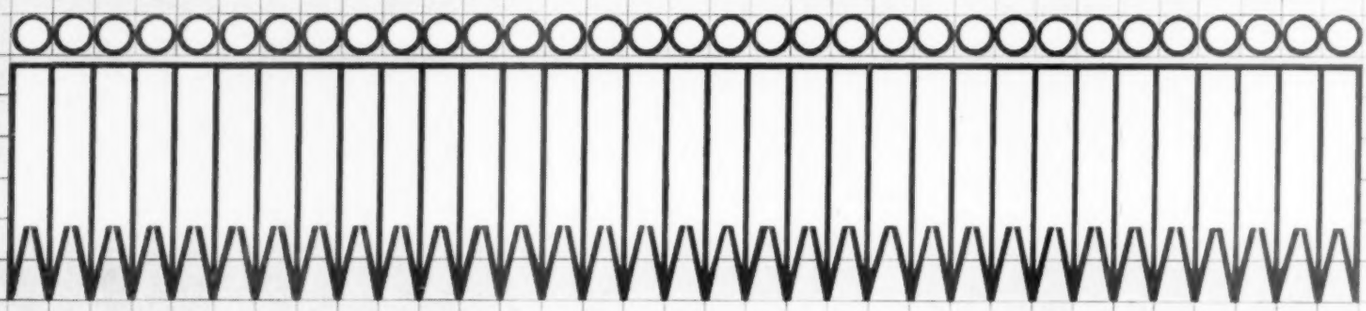
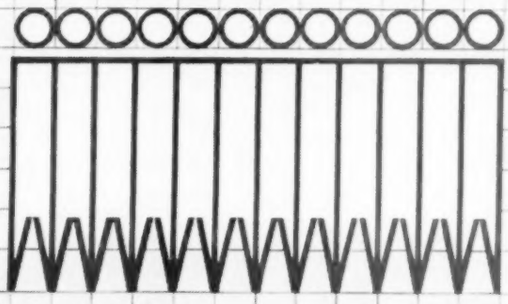
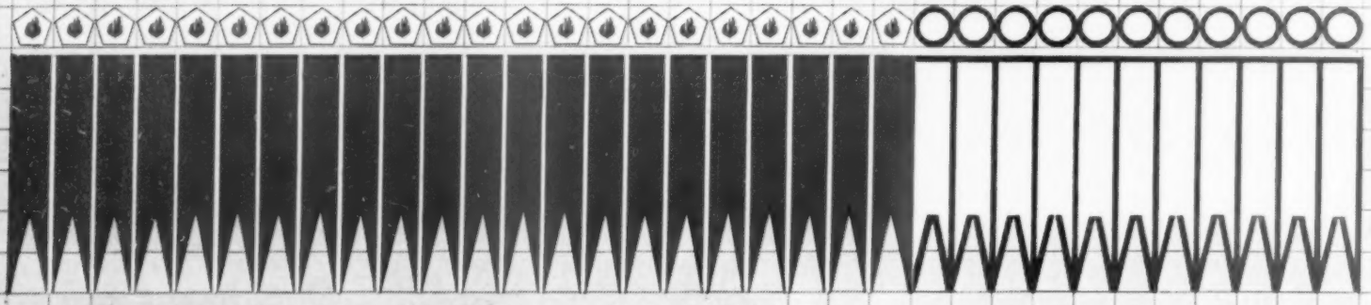


Of 45 arthritic patients
who were refractory
to other corticosteroids*





22 were successfully treated with **Decadron**[1,2]

1. Boland, E. W., and Headley, N. E.: Paper read before the Am. Rheum. Assoc., San Francisco, Calif., June 21, 1958.
2. Bunim, J. J., et al.: Paper read before the Am. Rheum. Assoc., San Francisco, Calif., June 21, 1958.

*Cortisone, prednisone and prednisolone.

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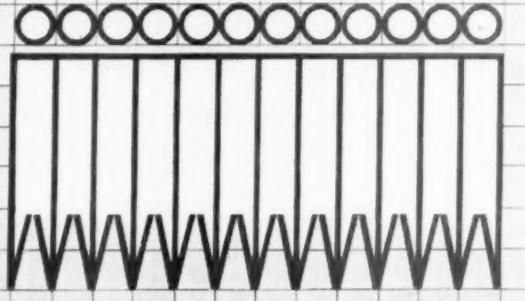

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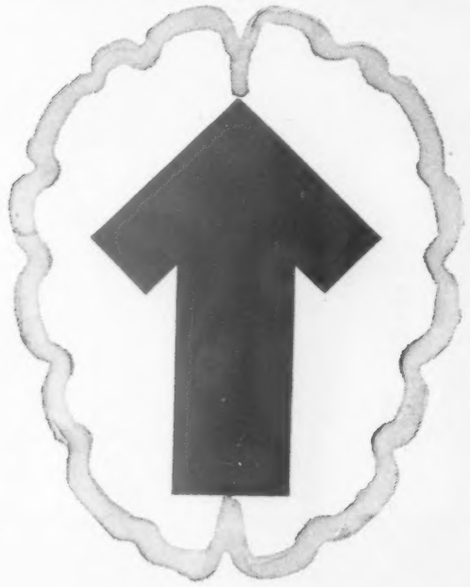
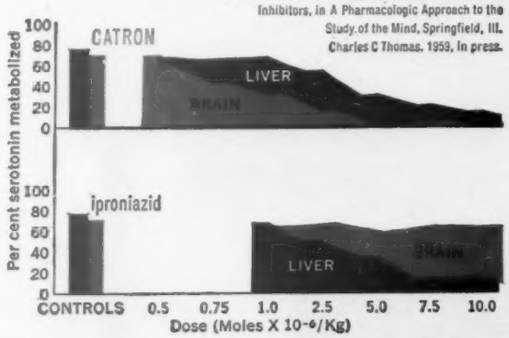


Elevates mood, brightens outlook by raising levels of mood-controlling neurohormones, serotonin and norepinephrine... at doses which have little or no effect on the liver.

Horita, A.: The Pharmacology of the Monoamine Oxidase Inhibitors, in A Pharmacologic Approach to the Study of the Mind, Springfield, Ill. Charles C Thomas, 1959, In press.



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gout

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By significantly increasing renal excretion of urate and thus lowering plasma uric acid, the new highly potent uricosuric agent ANTURAN strikes directly at the basic metabolic defect in gout.

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- Prevents formation of new tophi
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[1] Yu, T. F., Burns, J. J., and Gutman, A. B. Arth. & Rheumat. 1:532, 1958.

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withdrawn apathetic rejected gloomy
remorseful hopeless listless despairing
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WHEN THE WORDS
MEAN DEPRESSION,
THE TREATMENT IS

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β-phenylisopropyl hydrazine supplied as the hydrochloride

Important new psychoactive agent—acts selectively on the brain to brighten outlook, raise spirits, rebuild self-esteem, revitalize depressed patients.

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For detailed information, request Brochure No. 19, CATRON

Important New

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β-phenylisopropyl hydrazine supplied as the hydrochloride

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How to use this new drug:

CATRON Hydrochloride is a monoamine oxidase (MAO) inhibitor useful in the treatment of depression and of other disorders indicated below. It is recommended for use in carefully selected cases and in those patients who have not responded to the milder drugs.

ADMINISTRATION AND DOSAGE

Dosage of CATRON must be individualized according to each patient's response. The initial daily dose should not exceed 12 mg. and should be reduced as soon as the desired clinical effect is obtained. In severe depressions some clinicians desire rapid results and begin treatment with 24 mg. daily: this dosage should not be continued for more than a few days. A single daily dose in the morning is recommended. A continuous or interrupted schedule may be used, the latter during the maintenance period.

DEPRESSION (Endogenous, Reactive, Postpartum, Involutional and Depression Secondary to Schizophrenic or Neurotic Reaction): initially, 12 mg. once daily for approximately 2 weeks, or less if improvement appears. Dosage is then reduced to 6 mg. daily. As improvement continues, maintenance dosage of 6 mg. every other day or of 3 mg. daily often proves satisfactory. An interrupted dose schedule is recommended for long-term therapy.

ANGINA PECTORIS—3 to 6 mg. daily in most cases. Relief of pain and elevation of mood may be dramatic. Victims of angina pectoris who respond in this manner should be cautioned against overexertion induced by their sense of well-being.

RHEUMATOID ARTHRITIS (Adjunctive Therapy—in severely disabling forms, particularly when accompanied by depression): 9 to 12 mg. daily for 3 days, then 6 mg. daily, reducing further to 3 mg. daily on signs of improvement. If a conventional antiarthritic agent is used, lower doses of each are indicated.

CAUTION

Certain circumstances should be watched carefully when using CATRON.

DRUG POTENTIATION—The list of drugs which CATRON potentiates is not yet complete. CATRON should not be used concomitantly with any other drug unless, (a) it has been ascertained that the two drugs bear no qualitative relationship, or (b) potentiating action is being sought, as may be the case with tranquilizing drugs including reserpine and the phenothiazines, and with the amphetamines, barbiturates and hypotensive agents.

HYPOTENSIVE EFFECT—All normotensive patients receiving CATRON, but especially elderly patients, should be warned about the possibility of orthostatic hypotension during the initial period of higher dosage. In the few instances where this may occur, lowering of the dose will usually permit continuation of therapy.

COLOR VISION—A reversible red-green color defect has been reported in a few patients, chiefly hypertensives, on ex-

Psychoactive Agent

- Brightens mood, diminishes apathy and confusion, curbs symptoms of withdrawal, self-pity, inadequacy, despair.[1,2]
- Acts selectively on brain at doses having little or no effect on liver.[5-7]
- Valuable in depressions associated with chronic diseases such as angina pectoris,[8] severe rheumatoid arthritis.[9]

For detailed information, request Brochure No. 19, CATRON

tended therapy with CATRON. Discontinue the drug if such changes occur.

ANIMALS, NEUROLOGIC SIGNS—In toxicity studies with animals, a neurologic syndrome has been observed characterized by tremors, muscle rigidity and difficulty in locomotion. Although extensive clinical experience has not shown such reactions to be a problem in humans in recommended dosage, should a similar neurologic disturbance occur, the possibility of drug action should be considered.

SIDE EFFECTS—Major side effects requiring cessation of therapy are infrequent. Other side effects—constipation, delay in starting micturition, increased sweating, hyperreflexia, ankle edema, blurring of vision, dryness of the mouth—are usually readily controlled by lowering the dosage. Rash, observed in a few patients, cleared up rapidly upon discontinuing therapy.

WARNING: Pharmacologic studies show that with proper dosage CATRON will inhibit monoamine oxidase in the brain without influencing this enzyme in the liver. This is in contrast to previous inhibitors, which depress monoamine oxidase activity in the liver before affecting this enzyme in the brain.

Although the evidence suggests that serious life-threatening hepatitis seen with other MAO inhibitors should not occur with CATRON in the recommended dosage, it has been reported on rare occasion with dosages in excess of the recommended levels.

The Following Precautions are Recommended:

1. In all instances daily dose should not exceed 12 mg.
2. Reduce daily dose as soon as response is established, usually in a matter of 1 to 2 weeks.
3. Do not prescribe to a patient more than sixteen 6 mg. tablets or thirty-two 3 mg. tablets of CATRON at one time.
4. Patient should return for observation before additional CATRON is prescribed. For this reason, prescriptions for CATRON should be marked, "not refillable."
5. Perform regular liver function tests.
6. Do not use the drug in patients with a history of viral hepatitis or other liver abnormalities.

CATRON is the original brand of β-phenylisopropyl hydrazine. It is supplied as the hydrochloride in tablets of 3 mg. and 6 mg., bottles of 50.

(1) Agin, H. V.: The Use of JB-516 (CATRON) in Psychiatry, Conference on Amine Oxidase Inhibitors, New York Academy of Sciences, Nov. 20-22, 1958. (2) Bercel, N. A.: A Pharmacologic Approach to the Study of the Mind, Springfield, Ill., Charles C Thomas, 1959, in press. (3) Kinross-Wright, J.: Panel Discussion of Psychic Energizers, *ibid.* (4) Kinross-Wright, J.: Experience with JB-516 (CATRON) and Other Psychochemicals in Clinical Practice, Conference on Amine Oxidase Inhibitors, New York Academy of Sciences, Nov. 20-22, 1958. (5) Horita, A., and Parker, R. G.: Comparison of Monoamine Oxidase Inhibitory Effects of Iproniazid and Its Phenyl Congener, Proc. Soc. Exper. Biol. & Med. 99:617, 1958. (6) Horita, A.: *Beta*-Phenylisopropylhydrazine, A Monoamine Oxidase Inhibitor, Fed. Proc. 17:379, 1958. (7) Horita, A.: The Pharmacology of the Monoamine Oxidase Inhibitors, in A Pharmacologic Approach to the Study of the Mind, Springfield, Ill., Charles C Thomas, 1959, in press. (8) Kennamer, R., and Prinzmetal, M.: Treatment of Angina Pectoris with CATRON (JB-516), Am. J. Cardiol. 3:542, 1959. (9) Scherbel, A. L., and Harrison, J. W.: The Effects of Iproniazid and Some Other Amine Oxidase Inhibitors in Rheumatoid Arthritis, Conference on Amine Oxidase Inhibitors, New York Academy of Sciences, Nov. 20-22, 1958.

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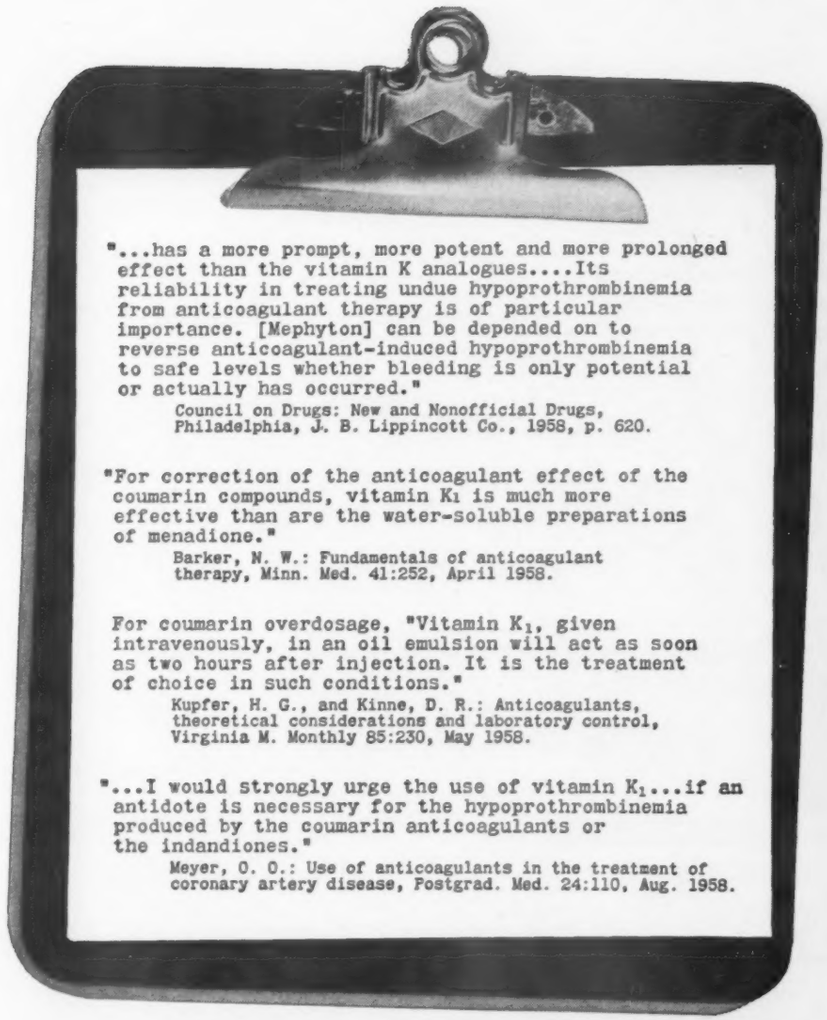
Consensus:
The preferred antidote for anticoagulant-induced hypoprothrombinemia is 'Mephyton' (vitamin K_1).



"...has a more prompt, more potent and more prolonged effect than the vitamin K analogues....Its reliability in treating undue hypoprothrombinemia from anticoagulant therapy is of particular importance. [Mephyton] can be depended on to reverse anticoagulant-induced hypoprothrombinemia to safe levels whether bleeding is only potential or actually has occurred."

Council on Drugs: New and Nonofficial Drugs, Philadelphia, J. B. Lippincott Co., 1958, p. 620.

"For correction of the anticoagulant effect of the coumarin compounds, vitamin K_1 is much more effective than are the water-soluble preparations of menadione."

Barker, N. W.: Fundamentals of anticoagulant therapy, Minn. Med. 41:252, April 1958.

For coumarin overdosage, "Vitamin K_1, given intravenously, in an oil emulsion will act as soon as two hours after injection. It is the treatment of choice in such conditions."

Kupfer, H. G., and Kinne, D. R.: Anticoagulants, theoretical considerations and laboratory control, Virginia M. Monthly 85:230, May 1958.

"...I would strongly urge the use of vitamin K_1...if an antidote is necessary for the hypoprothrombinemia produced by the coumarin anticoagulants or the indandiones."

Meyer, O. O.: Use of anticoagulants in the treatment of coronary artery disease, Postgrad. Med. 24:110, Aug. 1958.

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Dosage: Orally, to modify anticoagulant effects: 5 to 10 mg. initially; 15 to 25 mg. for more vigorous action. Intravenously, for anticoagulant-induced bleeding emergencies, 10 to 50 mg.; may be repeated as indicated by prothrombin time response. (Some clinicians advise their patients to keep a supply of tablets on hand at all times; if gross bleeding occurs, the patients are instructed to take 10 mg. and phone the doctor.)

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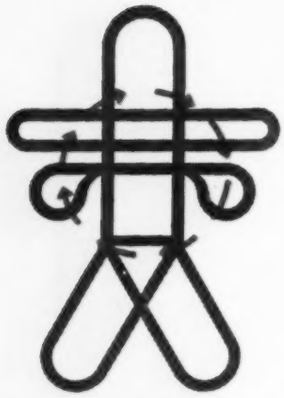

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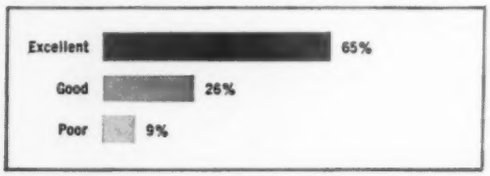

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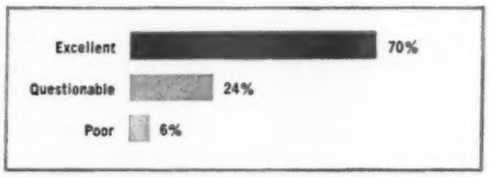

*Adapted from Singher, H. O., and Chapple, R. V.[5]

SAFETY—In recommended dosage ACTASE does not interfere with blood coagulation.

ADMINISTRATION—ACTASE is administered by intravenous infusion over a 2-hour period. ACTASE is most effective when given soon after the formation of the clot. The concomitant use of anticoagulants is recommended. A temperature rise (1° to 5.2° F.) may occur with ACTASE. This may be ameliorated with conventional antipyretic therapy.

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REFERENCES: (1) Astrup, T.: Lancet *2*:565 (Sept. 15) 1956. (2) Cliffton, E. E.: Ann. New York Acad. Sc. *68*:209 (Aug. 30) 1957. (3) Cliffton, E. E.: J. Am. Geriatrics Soc. *6*:118, 1958. (4) Sussman, B. J., and Fitch, T. S. P.: J.A.M.A. *167*:1705 (Aug. 2) 1958. (5) Singher, H. O., and Chapple, R. V.: Clin. Med. *6*:439 (March) 1959.

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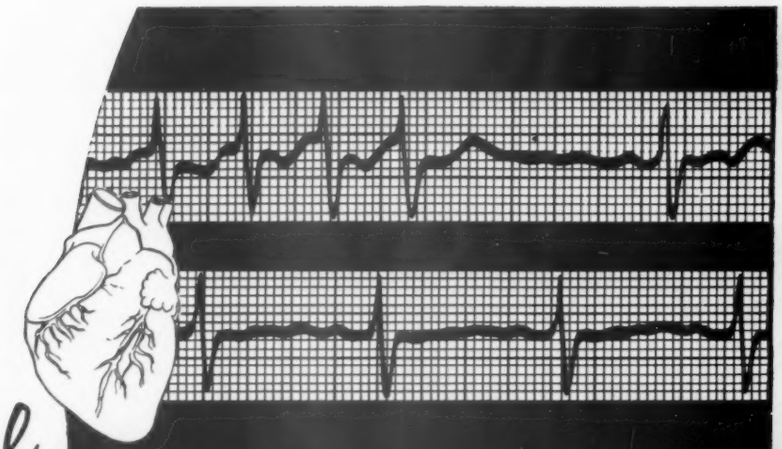

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PAGE 867

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1. Bellet, S., Finkelstein, D., and Gilmore, H.: A.M.A. Archives Internal Med. 100:750, 1957.

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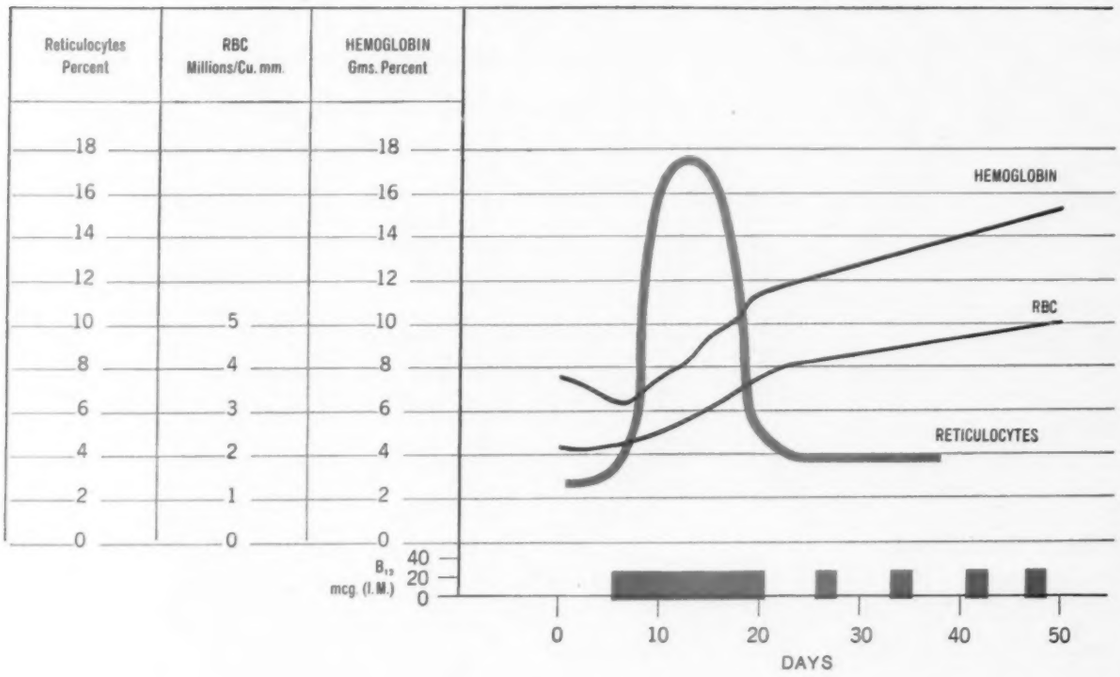

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Vitamin B_{12} – one of the most potent biological factors known – is vital to basic metabolic functions, to normal formation of red blood cells and other formed elements of the blood, and to the functional integrity of myelinated fibers in the spinal cord and brain, as well as to the healthy condition of gastric and oral mucosa.

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Vitamin B_{12} is the therapeutic agent of choice in pernicious anemia, is effective in certain megaloblastic macrocytic anemias, and contributes to recovery or clinical improvement in a variety of neurological, liver and skin disorders.

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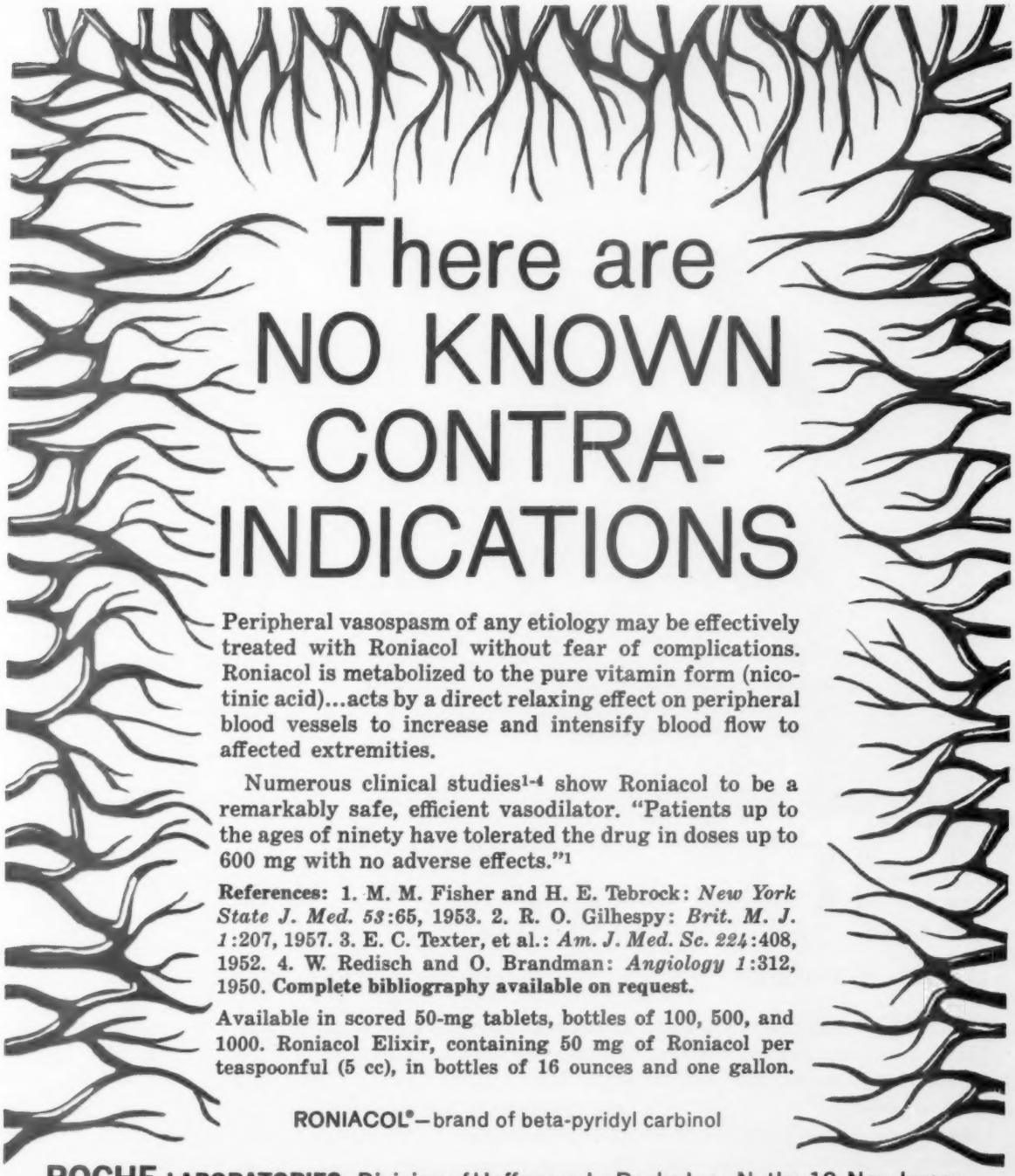

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References: 1. M. M. Fisher and H. E. Tebrock: *New York State J. Med. 53*:65, 1953. 2. R. O. Gilhespy: *Brit. M. J. 1*:207, 1957. 3. E. C. Texter, et al.: *Am. J. Med. Sc. 224*:408, 1952. 4. W. Redisch and O. Brandman: *Angiology 1*:312, 1950. **Complete bibliography available on request.**

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Butazolidin®

(phenylbutazone GEIGY)

tablets · alka capsules — **potent · nonhormonal · anti-inflammatory agent**

BUTAZOLIDIN tablets or the Alka capsules are **equally effective but individually adaptable in a wide range of arthritic disorders.**

Recent clinical reports continue to justify the selection of Butazolidin for rapid relief of pain, increased mobility, and early resolution of inflammation.

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References: 1. Graham, W.: Canad. M. A. J. 79:634 (Oct. 15) 1958. 2. Robins, H. M.; Lockie, L. M.; Norcross, B.; Latona, S., and Riordan, D. J.: Am. Pract. Digest Treat. 8:1758, 1957. 3. Kuzell, W. C.; Schaffarzick, R. W.; Naugler, W. E., and Champlin, B. M.: New England J. Med. 256:388, 1957.

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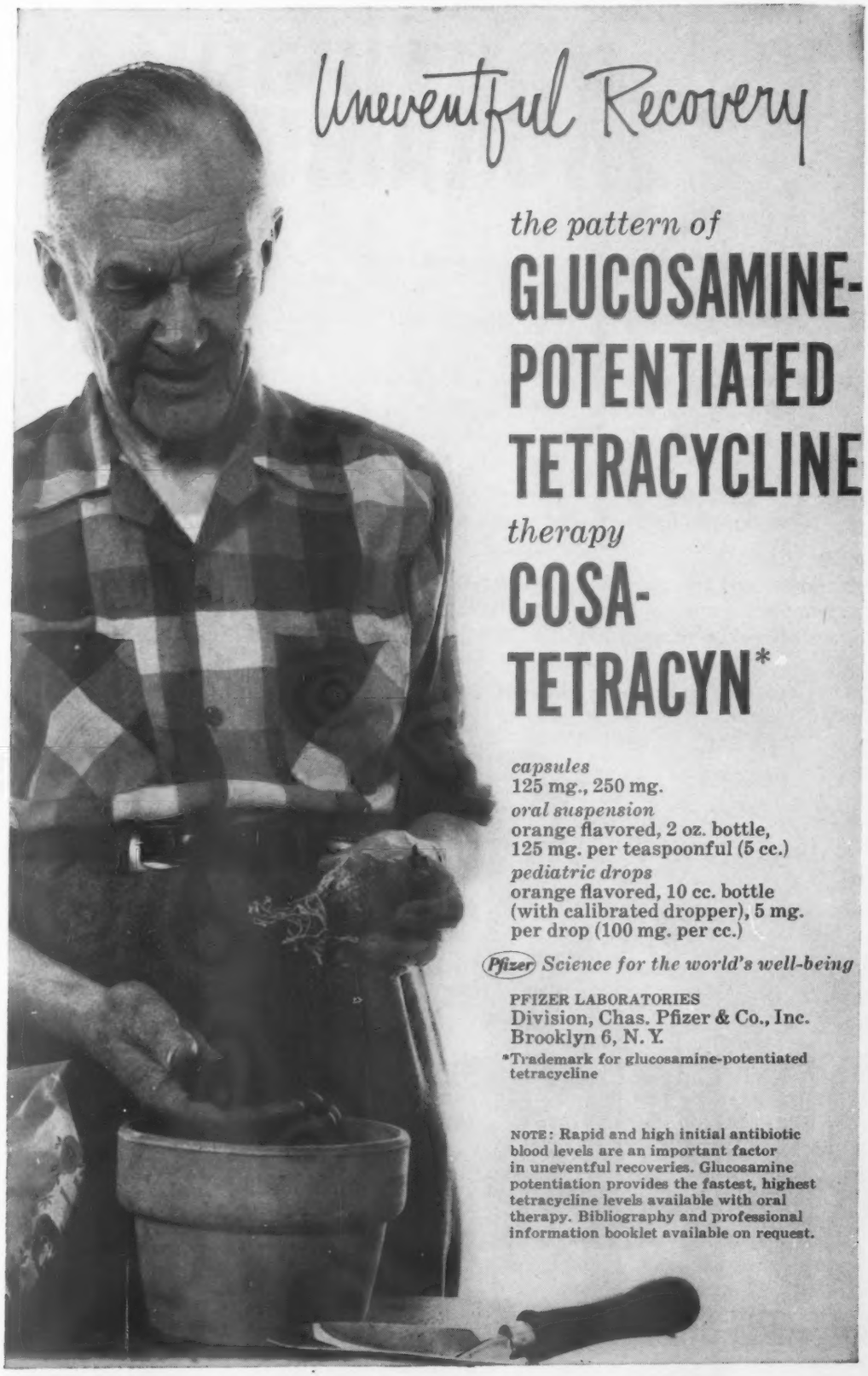

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orange flavored, 2 oz. bottle,
125 mg. per teaspoonful (5 cc.)
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orange flavored, 10 cc. bottle
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Rapid action	rate of absorption faster than menadione or derivatives... more potent and lasting effects.
Wide margin of safety	substantially safer than vitamin-K analogues—no kernicterus reported.
Versatility of administration	capsules for oral use...fine aqueous dispersion for parenteral administration.
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Prophylactically and therapeutically, Konakion is indicated in obstetrics to prevent or control neonatal hemorrhage, to minimize excessive bleeding in surgery, to offset anticoagulant overdosage, and whenever vitamin-K utilization is impaired.

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2. Richardson, M. E.: J. Am. Osteop. A. 57:562 (May) 1958.
3. Mason, M. L.: Northwest Med. 57:1439 (Nov.) 1958.

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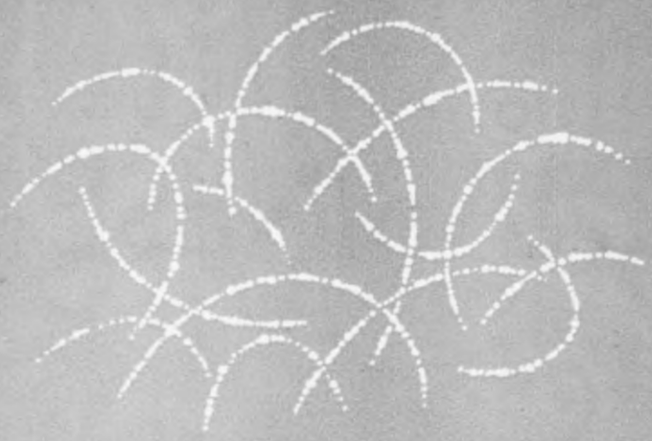

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brand of chlorpropamide

tablets / once-a-day dosage

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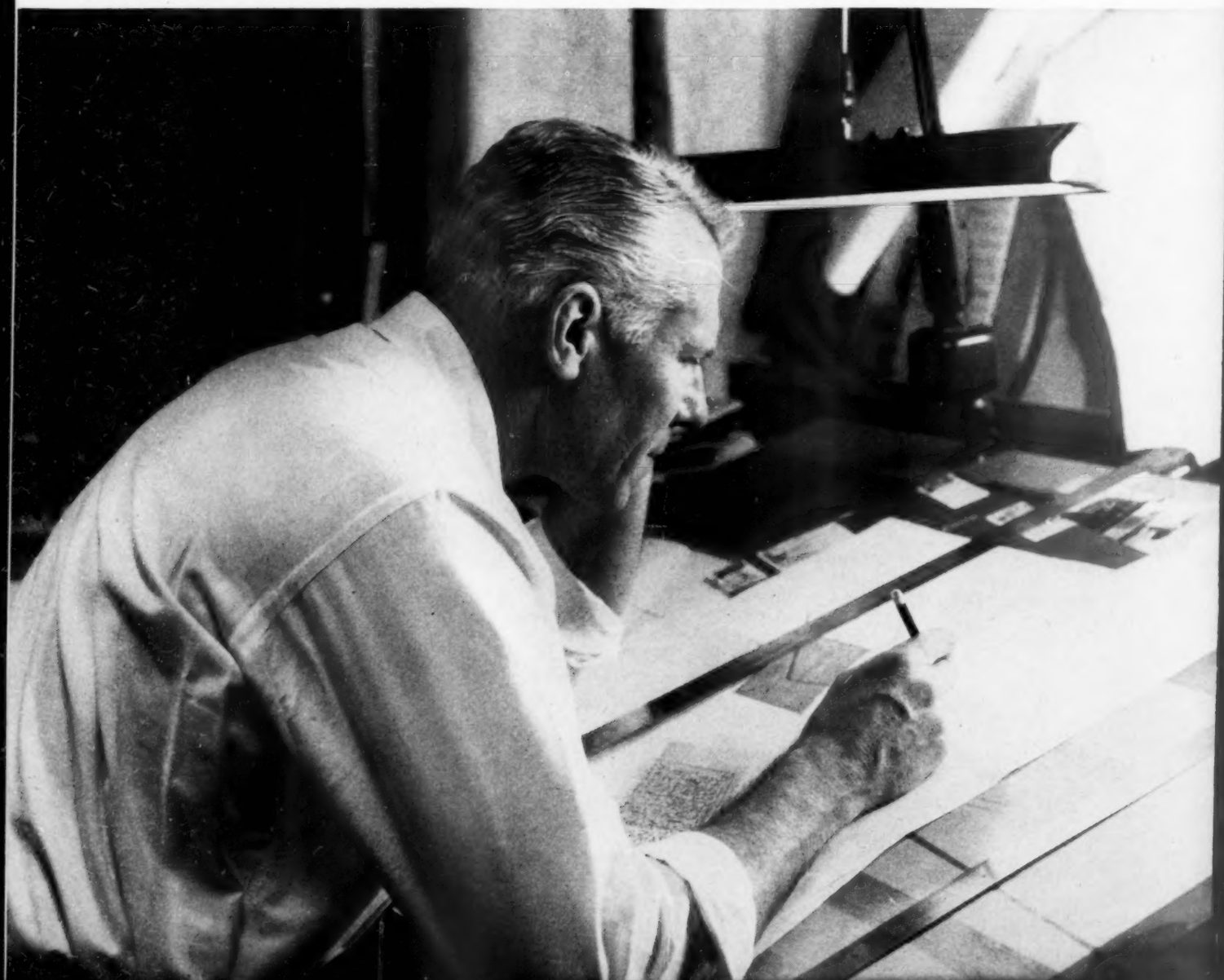
1. Dobson, H., et al.: Ann. New York Acad. Sc. 74:940, 1959. 2. Greenhouse, B.: Ibid., p. 643. 3. Forsham, P. H.; Magid, G. J., and Dorosin, D. E.: Ibid., p. 672. 4. Beaser, S. B.: Ibid., p. 701; New England J. Med. 259:573, 1958. 5. Blöch, J., and Lenhardt, A.: Ann. New York Acad. Sc. 74:954, 1959. 6. O'Driscoll, B. J.: Lancet 2:749, 1958. 7 Hadley, W. B.; Khachadurian, A., and Marble, A.: Ann. New York Acad. Sc. 74:621, 1959. 8. Duncan G. G.; Schless, G. L., and Demeshkieh, M. M. A.: Ibid., p. 717. 9. Handelsman, M. B.; Levitt. L., and Calabretta, M. F.: Ibid., p. 632. 10. Hills, A. G., and Abelove, W. A.: Ibid., p. 845. 11. Drey, N. W., et al.: Ibid., p. 962.



now potent tranquilizer therapy is safer than ever

Virtual freedom of Mellaril from major toxic effects is due to greater specificity of tranquilizing action – divorced from such "diffuse" effects as anti-emetic action.

MELLARIL is virtually free of such toxic effects as

- jaundice
- Parkinsonism
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"Thioridazine [MELLARIL] is as effective as the best available phenothiazine, but with appreciably less toxic effects than those demonstrated with other phenothiazines. . . . This drug appears to represent a major addition to the safe and effective treatment of a wide range of psychological disturbances seen daily in the clinics or by the general practitioner."

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THIORIDAZINE HCl

specific, effective tranquilizer • safer at all dosage levels

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"The most striking aspect of thioridazine [Mellaril] therapy is the poverty of side effects.... In its lack of side effects and low toxicity, it is superior to all other tranquilizing drugs tested. For this reason also it is well tolerated by patients, particularly those who are not hospitalized and who frequently discontinue their medication because of dizziness, sleepiness, increased tension or parkinsonism with other drugs."[2]

NEGLIGIBLE SIDE EFFECTS

"Side effects were negligible at all dosage levels: no incidence of parkinsonism or other extrapyramidal symptoms. Minimal sedation, on the whole lower than with other tranquilizing agents. No alteration in liver function, urine or blood. No photosensitivity. Patient acceptability was exceptional: lack of drowsiness, lethargy or 'washed out' feeling, permitted patients to carry on normal everyday activities. Orthostatic hypotension was absent. The initial 'keyed up' tense feeling common to other drugs of this type was absent.... Patients forced to interrupt treatment with other phenothiazine derivatives because of parkinsonism or other extrapyramidal symptoms were able to continue therapy with thioridazine without appearance of parkinsonism."[3]

SINGULARLY FREE OF SIDE EFFECTS

"The extrapyramidal syndrome was not encountered in any of its forms. Dizziness and sleepiness responded to a reduction in dosage. Other side effects did not occur.... It is singularly free from the side effects ordinarily seen with these [phenothiazine] compounds."[4]

ABSENCE OF SIGNIFICANT SIDE EFFECTS

"None of the following toxic effects, so common after administration of the phenothiazines, was present during the period of Thioridazine administration: Parkinsonism or Parkinson-like symptoms, photosensitivity, orthostatic hypotension, bone-marrow depression."[1]

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"Side effects such as extrapyramidal activity, jaundice and photosensitivity have not been observed in patients treated with Thioridazine [Mellaril]. Extrapyramidal side effects produced by other phenothiazines have disappeared promptly with no deterioration in the behavioral response when these patients have been shifted to Thioridazine."[5]

NO JAUNDICE

"No allergic reactions were observed such as skin eruptions, jaundice or agranulocytosis. Central nervous system toxicity, as manifested by extrapyramidal effects, seizures, and excitement did not occur despite the use of high doses (up to 2000 mg.) of the drug."[6]

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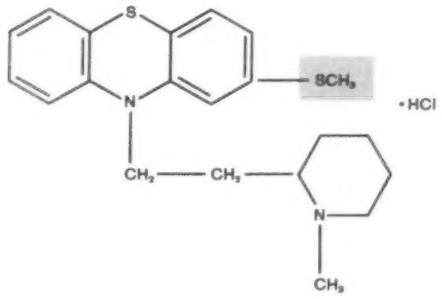
THIORIDAZINE HCl

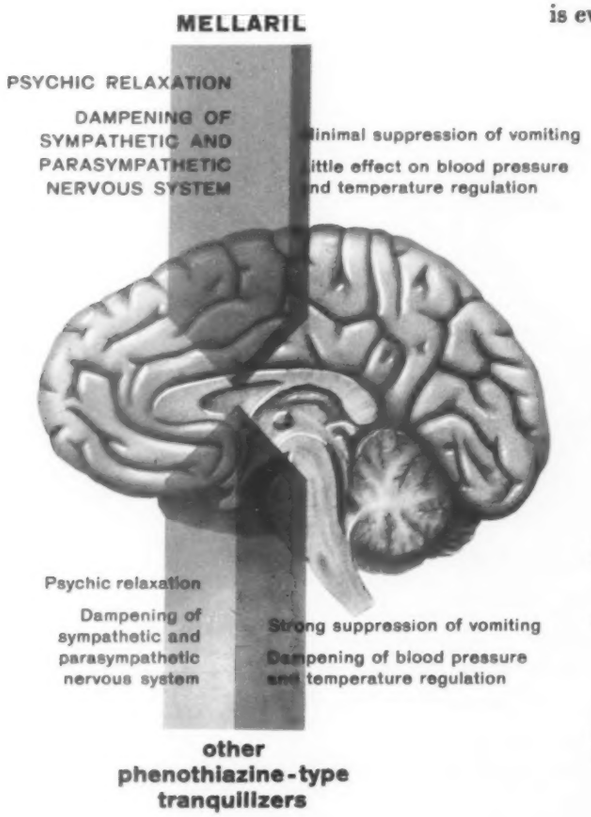
specific, effective tranquilizer • safer at all dosage levels

a new advance in tranquilization:
greater specificity of tranquilizing action plus fewer side effects



Of 109 phenothiazines synthesized by Sandoz, Mellaril was selected as the most promising on the basis of extensive evaluation. The presence of a thiomethyl radical ($S\text{-}CH_3$) in the position conventionally occupied by a halogen in other phenothiazines is unique and could be responsible for the relative absence of side effects and greater specificity of psychotherapeutic action. This is shown clinically by:

1 **A specificity of action on certain brain sites in contrast to the more generalized or "diffuse" action of other phenothiazines. This is evidenced by a lack of appreciable anti-emetic effect.**


MELLARIL
PSYCHIC RELAXATION
DAMPENING OF SYMPATHETIC AND PARASYMPATHETIC NERVOUS SYSTEM
Minimal suppression of vomiting
Little effect on blood pressure and temperature regulation
Psychic relaxation
Dampening of sympathetic and parasympathetic nervous system
Strong suppression of vomiting
Dampening of blood pressure and temperature regulation
other phenothiazine-type tranquilizers

2 **Less "spill-over" action to other brain areas — hence, absence of undue sedation, drowsiness or autonomic nervous system disturbances.**

3 **A notable absence of extrapyramidal stimulation.**

4 **Lack of impairment of patient's normal drive and energy, while achieving psychomotor control in mental and emotional disorders.**

5 **Virtual freedom from toxic effects — jaundice, photosensitivity, skin eruptions, disturbed body temperature regulation, blood forming disorders have been absent in reports currently available.**

These properties add up to a greater margin of safety in general office practice, in ambulatory psychiatric out-patient clinics, and in hospitalized patients.

Mellaril®

THIORIDAZINE HCl

specific, effective tranquilizer • safer at all dosage levels

excellent clinical response

In office practice and in hospitalized patients, Mellaril has proved highly useful for a wide variety of major and minor emotional disorders (such as anxiety, tension, apprehension, alcoholism, agitated psychoneurosis, agitated psychotic states, etc.).

EXTREMELY SATISFACTORY ". . . produced extremely satisfactory results in the broad therapeutic range represented in this series."[3]

POTENT AGENT ". . . appears to be a potent agent in the symptomatic management of a variety of psychiatric states."[4]

MAJOR ADDITION TO THERAPEUTICS "This drug appears to represent a major addition to the safe and effective treatment of a wide range of psychological disturbances seen daily in the clinics or by the general practitioner."[1]

AN ACTIVE AGENT "Thioridazine is an active therapeutic agent. . . . It is effective in a variety of psychiatric disorders, including schizophrenic reactions. . . . The drug is particularly advantageous for a group of schizophrenic patients who are sometimes made worse by other phenothiazine derivatives or Rauwolfia alkaloids. It should also be suitable for treating patients with psychoneuroses and chronic brain syndrome."[6]

EVEN IN VERY SEVERE CASES "Of the 152 patients treated 25 have been released and they have not suffered a relapse. This proportion is significant if we stop to consider that we are dealing only with acute cases which had been considered hopeless and obviously destined to finish their days in an asylum."[7]

EXCELLENT THERAPEUTIC RESPONSE "Patients with emotional tensions resulting from the stress and strain of life . . . were treated with Mellaril at the dosage level of 10 mg. three times daily. In 94 such patients, 83 obtained an excellent therapeutic response."[8]

Mellaril®

THIORIDAZINE HCl

specific, effective tranquilizer • safer at all dosage levels

"... extremely satisfactory results ..." in a clinical spectrum ranging from minor nervous disorders to severe psychotic disturbances[3]

RESULTS WITH MELLARIL IN 194 PATIENTS[3]

ACUTE PSYCHOTICS	CHRONIC PSYCHOTICS	NEUROTICS
83% satisfactory effect	68% satisfactory effect	57% satisfactory effect
Some cases had complete remission of symptoms. Most were able to return home to useful occupations.	Relief of symptoms in cases permitted easier management and a return to a more or less useful life.	Some cases, complete relief of symptoms. Other cases, partial relief of symptoms.

RESULTS WITH MELLARIL IN PATIENTS PREVIOUSLY TREATED WITH OTHER TRANQUILIZERS[3]

DIAGNOSTIC CATEGORY	IMPROVED %	VERY SATISFACTORY %	SATISFACTORY %	UNSATISFACTORY %
SCHIZOPHRENIA				
Acute	89	61	28	11
Chronic paranoid	84.2	31.6	52.6	15.8
Chronic, other	73.9	21.7	52.2	26.1
Residual	57.1	9.5	47.6	42.9
CHRONIC BRAIN SYNDROME	66.6	33.3	33.3	33.3
CHRONIC PSYCHONEUROSIS	62.5	12.5	50	37.5
CHRONIC PSYCHOSOMATIC DISORDERS	75	25	50	25

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a guide to administration and dosage

Dosage ranges from 10 mg. three or four times a day in milder situations to 25 mg. three or four times a day for more disturbed patients. In ambulatory psychiatric out-patients, dosages of 50 to 100 mg. three or four times a day have been found adequate. For severely disturbed hospitalized psychotics, dosages of 200 to 300 mg. three times a day may be administered.

Dosage must be individualized according to the condition and degree of response. In all cases, the smallest effective dosage should be determined for each patient.

INDICATION	USUAL STARTING DOSE	TOTAL DAILY DOSAGE RANGE
	ADULTS	
Mental and Emotional Disturbances:		
MILD—where anxiety, apprehension and tension are present	10 mg. t.i.d.	20-60 mg.
MODERATE—where agitation exists in psychoneurosis, alcoholism, intractable pain, senility, etc.	25 mg. t.i.d.	50-200 mg.
SEVERE—in agitated psychotic states as schizophrenia, manic depressive, toxic psychoses, etc.:		
Ambulatory	100 mg. t.i.d.	200-400 mg.
Hospitalized	100 mg. t.i.d.	200-800 mg.
	CHILDREN	
BEHAVIOR PROBLEMS IN CHILDREN	10 mg. t.i.d.	20-40 mg.

PRECAUTIONS: Although possessing a unique structure and a selectivity of action which broadens its therapeutic ratio, the physician should be alert to the possibility of untoward reactions in certain susceptible individuals. In particular, he should watch for potential hemopoietic depression, jaundice or orthostatic hypotension. As with other phenothiazines, Mellaril is contraindicated in severely depressed or comatose states from any cause.

SUPPLIED: MELLARIL Tablets, 10 mg., 25 mg., 100 mg. Bottles of 100.

1. Ostfeld, A. M.: Scientific Exhibit, American Academy of General Practice, San Francisco, April 6-9, 1959. 2. Kinross-Wright, V. J.: Lecture, Clinical Meeting, American Medical Association, Minneapolis, Dec. 4, 1958. 3. Kinross-Wright, V. J.: Scientific Exhibit, Clinical Meeting, American Medical Association, Minneapolis, Dec. 2-5, 1958. 4. Cohen, S.: TP-21, a new phenothiazine, Am. J. Psychiat. *115*:358, Oct. 1958. 5. Glueck, B.: Scientific Exhibit, American Psychiatric Association, Philadelphia, April 27-May 1, 1959. 6. Hollister, L. E., and Macdonald, B. F.: Presented at California Medical Association; Section on Psychiatry, San Francisco, Feb. 25, 1959. 7. Remy, M.: Schweiz. med. Wchnschr. *88*:1221, Nov. 29, 1958. 8. Freed, S. C., in discussion on Thioridazine (Mellaril) in Psychiatric Patients, Hollister, L. E., and Macdonald, B. F., presented at California Medical Association; Section on Psychiatry, San Francisco, Feb. 25, 1959.

- controls neurotic and psychotic patients with anxiety, apprehension, nervous tension
- virtual absence of jaundice, parkinsonism, photosensitivity, dermatitis
- minimal sedation and drowsiness
- does not mask organic conditions such as brain tumors, intestinal obstruction, etc., because of lack of anti-emetic action
- increased specificity of action results in greater safety at all dosage levels

SANDOZ

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THIORIDAZINE HCl

specific, effective tranquilizer • safer at all dosage levels

Established Standard Therapy in Hypertension*

Rauwiloid

alseroxylon, 2 mg.

*Because

Rauwiloid provides effective Rauwolfia action virtually free from side effects... the smooth therapeutic efficacy of Rauwiloid is associated with significantly less toxicity than reserpine... and with a lower incidence of depression. Tolerance does not develop.

Rauwiloid is initial therapy for every hypertensive patient. ...Dosage adjustment is never a problem...

just two tablets at bedtime

After full effect one tablet suffices

•

When more potent drugs are needed, prescribe one of the *convenient single-tablet combinations*

Rauwiloid-Ventolin

alseroxylon 1 mg. and alkavervir 3 mg.

or

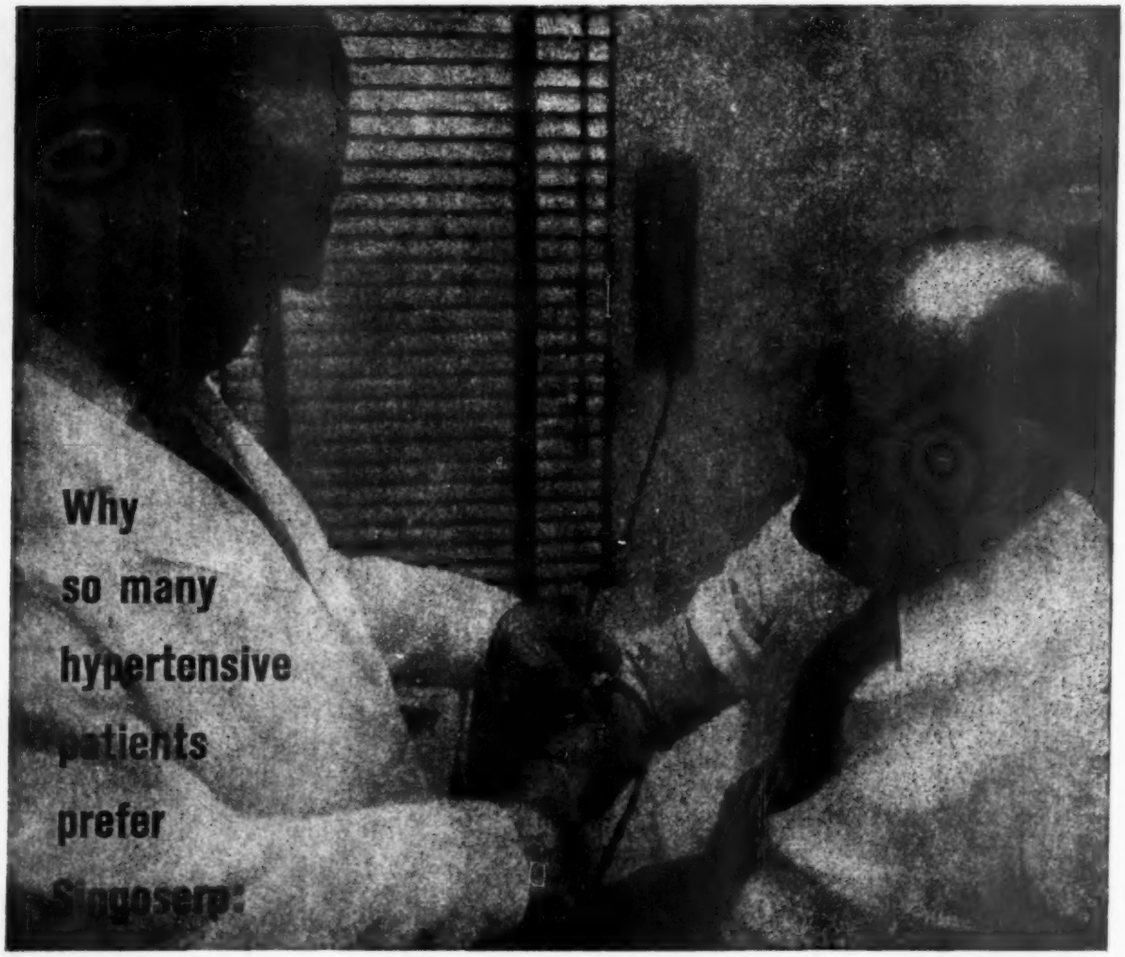
Rauwiloid-Hexamethonium

alseroxylon 1 mg. and hexamethonium chloride dihydrate 250 mg.

Many patients with severe hypertension can be maintained on Rauwiloid alone after desired blood pressure levels are reached with combination medication.

Riker

Northridge, California



It spares them from the usual rauwolfia side effects

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*Herrmann, G. R., Vogelpohl, E. B., Hejtmancik, M. R., and Wright, J. C.: J.A.M.A. 169:1609 (April 4) 1959.

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First drug to try in new hypertensive patients

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SUPPLIED: Singoserp *Tablets*, 1 mg. (white, scored); bottles of 100. Samples available on request. Write to CIBA, Box 277, Summit, N. J.

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